BIOSECURITY:

THE PATH TO RAISING PIGS WITHOUT ANTIBIOTICS?

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I remember.

- Sam Fender -

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LIST OF ABBREVIATIONS

A. pleuropneumoniae	*Actinobacillus pleuropneumoniae*
AACTING	Network on quantification of veterinary antimicrobial use at herd level and analysis, communication and benchmarking to improve responsible use
ADG	Average daily gain
ADKAR®	Awareness, Desire, Knowledge, Ability, Reinforcement
AGP	Antimicrobial growth promoter
AMCRA	Belgian knowledge center on antimicrobial use and resistance in animals
AMR	Antimicrobial resistance
AMU	Antimicrobial use
ASF	African swine fever
B. bronchiseptica	*Bordetella bronchiseptica*
B. hyodysenteriae	*Brachyspira hyodysenteriae*
BD100	Behandeldagen met antibiotica per 100 dagen (number of treatment days with antibiotics in 100 days; percentage of treatment days with antibiotics)
BelVet-SAC	Belgian veterinary surveillance of antimicrobial consumption
BFS	Batch farrowing system
BW	Body weight
CIA	Critically important antimicrobials
DANMAP	Danish integrated antimicrobial resistance monitoring and research program
$DDDA_{bel}$	Belgian defined daily dose animal
DNA	Deoxyribonucleic acid
E. coli	*Escherichia coli*
E. rhusiopathiae	*Erysipelothrix rhusiopathiae*
EFSA	European Food Safety Authority

ELISA	Enzyme-linked immunosorbent assay
EMA	European Medicines Agency
ESVAC	European Surveillance of Veterinary Antimicrobial Consumption
EU	European Union
FAO	Food and Agriculture Organization of the United Nations
FAVV	Federaal Agentschap voor de Veiligheid van de Voedselketen (The Federal Agency for the Safety of the Food Chain)
FCR	Feed conversion ratio
FOD VVVL	Federale Overheidsdienst Volksgezondheid Veiligheid van de Voedselketen en Leefmilieu (The Federal Public Service Health, Food Chain Safety and Environment)
G. parasuis	*Glaeserella parasuis*
GDU	Gilt development unit
IU	International units
L. intracellularis	*Lawsonia intracellularis*
LA_{bel}	Long-acting factor for Belgium
M. hyopneumoniae	*Mycoplasma hyopneumoniae*
NSF	National Sanitation Foundation
P. multocida	*Pasteurella multocida*
PCR	Polymerase chain reaction
PCU	Population correction unit
PCV-2	Porcine circovirus type 2
PLF	Precision livestock farming
PM	Particulate matter
PMWS	Post-weaning multisystemic wasting syndrome
PPV	Porcine parvovirus

PRRSV	Porcine reproductive and respiratory syndrome virus
PRVA	Porcine rotavirus type A
QS	Qualitätssicherung (quality scheme for safe food)
ROI	Return on investment
RWA	Raised without antibiotics
S. hyicus	*Staphylococcus hyicus*
S. suis	*Streptococcus suis*
SD	Standard deviation
SIV	Swine influenza virus
SPC	Summary of product characteristics
SPF	Specific pathogen free
spp.	Species
WHO	World Health Organization
WOAH	World Organization for Animal Health
WTP	Willingness to pay

CHAPTER 1 | GENERAL INTRODUCTION

1. INTENSIVE PIG PRODUCTION

1.1 PIG PRODUCTION WORLDWIDE AND IN EUROPE

Livestock production contributes significantly to the global food production. Growth of the global population and increasing wealth lead to a growing demand for animal-source foods. Traditional small-scale or mixed farms were not able to fully meet these demands. Therefore, livestock production shifted towards a more intensive large-scale production of a single product, also called the livestock revolution (Steinfeld, 2004).

Worldwide, pigs are one of the most commonly raised animals for meat production (Robinson et al., 2011). Pigs have a short generation interval and a low feed conversion ratio, which makes them very suitable for meeting the rising need for animal protein. Pigs are the primary source of protein for millions of people in different cultures and geographical regions. In 2021, 122.4 million tons of pig meat were produced worldwide. Some countries or regions are responsible for the majority of pork production, namely China (44 %), the European Union (19 %), The United States of America (10 %), Brazil (4 %), Russia (4 %), Vietnam (3 %), and Canada (2 %). The remaining 14 % of pork is produced by other countries (Food and Agriculture Organization of the United Nations (FAO), 2022).

In Europe, pig production is an important agricultural sector, with the European Union (EU) being the second largest producer of pork worldwide (European Commission, 2023a). In 2021, there were 142 million pigs in the EU. Two thirds of these were held in just a few of the EU Member States, namely Spain (24.3 %), Germany (16.8 %), Denmark (9.3 %), France (9.1 %), and the Netherlands (7.7 %). The remaining 32.8 % of the pigs were raised in other EU Member States. In 2021, 23.4 million tons of pig meat were produced in the EU, an increase of 9.4 % compared to 2006, with the main increase occurring after 2013. This increase in pork production was achieved even though the total number of pigs decreased since then, which can be explained by improved genetics and management practices, leading to a more efficient meat production. Also for the EU pork production, a few countries were responsible for the majority of the meat production, namely Spain (22.1 %), Germany (21.2 %), France (9.4 %), Poland (8.4 %), and Denmark (7.4 %). The remaining 31.4 % of pork originated from other EU Member States (European Commission, 2022). The EU is the biggest exporter of pork (products), with an export of about 13 % of the total pork production, mainly to East Asia and in particular to China (European Commission, 2023b).

1.2 PIG PRODUCTION IN BELGIUM

Over the past two decades, the number of pig farms in Belgium has shown a decreasing trend, while the number of pigs per farm is increasing. The total number of pigs used to be quite stable, but since 2004 there has been a large decrease in the number of sows and a slight decrease in the total number of pigs. In 2022, there were 5.7 million pigs in Belgium, including 1.6 million nursery pigs, 3.8 million fattening pigs, and 367 866 breeding pigs. These pigs were housed on 4108 farms. The focus of pig production in Belgium is in Flanders, where 94 % of the animals and 85 % of the farms are located. Moreover, 53 % of all Belgian pigs are housed in one province, namely West Flanders (Figure 1) (StatBel, 2022b; Statistiek Vlaanderen, 2023). In 2022, 10.5 million pigs were slaughtered in Belgium (StatBel, 2022c). Belgium has a self-sufficiency rate of 239 %, meaning that pork export is very important. In 2022, 1.1 million tons of pork were produced, and 75 % of pork (products) were exported (StatBel, 2022a).

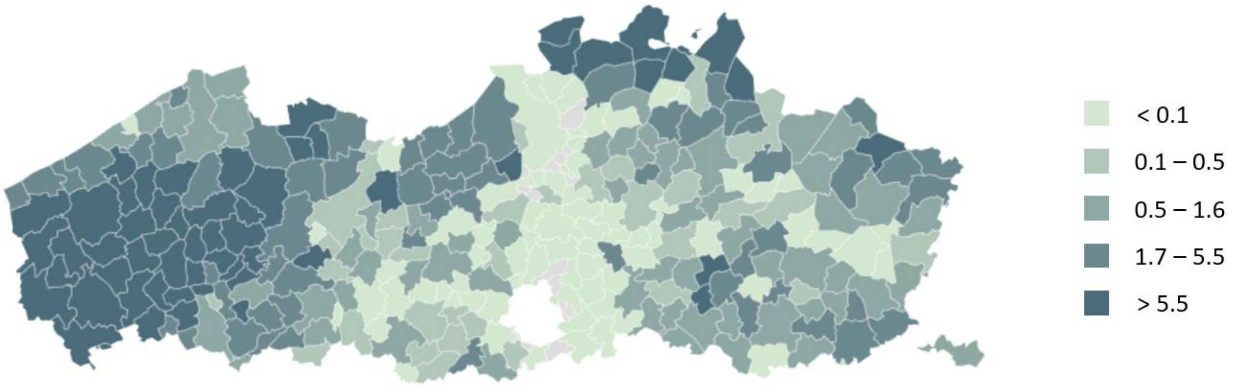

Figure 1. Pig density in the municipalities of Flanders in 2021, in number of pigs per hectare (Statistiek Vlaanderen, 2023).

2. ANTIMICROBIALS

2.1 ORIGIN OF ANTIMICROBIALS

Antimicrobials are a group of drugs that are used to treat infections caused by bacteria, viruses, fungi, and other micro-organisms. Their history dates back to the early 20th century, when the first antibiotic, penicillin, was discovered. In 1928, Alexander Fleming found the fungus *Penicillium notatum* on an agar-medium plate containing staphylococci and noticed that the bacteria around this fungus had disappeared. Apparently, this fungus secreted a product capable of killing bacteria. The purification of the substance was completed only more than a decade later by two scientists, Florey and Chain. After this, penicillin could be used as a therapeutic drug. During World War II, penicillin was mass-produced and used to treat soldiers against bacterial infections and septicemia. The discovery of penicillin, also referred to as the miracle drug, revolutionized medicine and paved the way for the development of other antimicrobial drugs. In 1945, Fleming, Florey, and Chain, were rewarded for their work and shared the Nobel Prize in Physiology/Medicine (Aminov, 2010).

The period between the 1950s and 1970s is referred to as the golden era of antimicrobial discovery; and many new classes of antimicrobials were discovered and developed for therapeutic use. This golden era came to an end in the 1970s, when the pace of discovering new antimicrobials slowed down and the problem of antimicrobial resistance (AMR) began to emerge. Since the 1970s, no new antimicrobials were discovered and currently, the development of new drugs is based on modification of existing antimicrobials (Aminov, 2010). Throughout this thesis, the term antimicrobials will be used to describe agents with antibacterial activity.

2.2 ANTIMICROBIAL USE IN INTENSIVE LIVESTOCK PRODUCTION

In the 1950s, antimicrobials were introduced in livestock production to meet the growing demand for animal protein. The livestock revolution was accompanied by higher animal stocking densities, resulting in efficient pathogen transmission between live animals. There are mainly three different ways for using antimicrobials in pig production. Curative or therapeutic medication implies that antimicrobials are given to diseased animals preferably after diagnosis of a bacterial infection. Metaphylactic medication refers to giving antimicrobials to a group of animals after diagnosis of infection and/or clinical disease in a part of a group to prevent the spread of infection to animals in close contact. Prophylactic or preventive medication is the administration of antimicrobials to an animal or a group of animals without the presence of clinical signs. It is practiced with the aim to prevent the onset of disease, e.g. in periods of stress such as weaning (Aarestrup, 2005).

Prophylactic or preventive administration of antimicrobials is considered to be imprudent. Furthermore, in December of 2018, the new EU regulation on Veterinary Medicinal Products was communicated and it came into force in January 2022. One of the main goals of this new regulation was to strengthen the EU response to fight antimicrobial resistance and it was determined that prophylactic use of antimicrobials should only be used in exceptional cases for administration to individual animals when the risk of infection is very high or the consequences are likely to be severe. Furthermore, the veterinarian should be able to justify the prescription of antimicrobials, especially in the case of metaphylactic and prophylactic use. This new regulation bans the prophylactic use of antimicrobials in groups of animals (European Council Regulation, 2018).

In the past, antimicrobials were administered to animals to improve growth; they were then referred to as antimicrobial growth promoters (AGPs). Previous studies have shown the impact of antimicrobials in feed on production. However, recent studies have concluded that the current productivity benefits of using AGPs in feed have diminished due to the adoption of modern production and management practices. The use of AGPs gradually phased out in Europe in 2000 and in 2003 the EU decided to ban all AGPs in EU Member States by 2006 (European Council Regulation, 2003). Initially, there was a slight and short shift to more therapeutic antimicrobial use (AMU) in Denmark and The Netherlands, but this increase turned out to be only temporary and was non-existent in other countries (Dewulf et al., 2022). Although AGPs were banned in the EU, 25 % of the countries worldwide (mainly countries in the Americas) were still using AGPs in 2020. In some countries, AGPs were no longer used, even though there was no specific legislation for banning AGPs. In contrast, there were also countries using antimicrobials as growth promoters within a regulatory framework, indicating that enforcement of legislation is needed (World Organization for Animal Health (WOAH), 2022).

2.3 ANTIMICROBIAL USE IN PIG PRODUCTION IN EUROPE AND BELGIUM

In this paragraph, different aspects of antimicrobial use in pig production will be described, namely the active ingredients, the administration routes and dosages, treatment indications, treatment age, and the risk factors. Also (monitoring of) AMU in the different European countries will be discussed, followed by a detailed description of AMU in Belgium.

There is a lot of variation in antimicrobials that are being used. For oral group medication, doxycycline, amoxicillin, trimethoprim-sulfonamides, and colistin are often used, while parenteral treatments often include long-acting amoxicillin (Callens et al., 2012; Dewulf et al., 2022; Jensen et al., 2012; Sjölund et al., 2015; Timmerman et al., 2006; van Rennings et al., 2015).

Antimicrobials can be administered parenterally, i.e. via injection, or orally, i.e. via feed or drinking water. The first one is mostly used for treatment of individual animals, while the latter is used for treatment of groups of animals. In some studies, parenteral treatments were most common (Sjölund et al., 2015), while in others oral treatments with antimicrobials were prevailing (Callens et al., 2012; Chauvin et al., 2002; Sarrazin et al., 2019; Sjölund et al., 2016). A recent study showed that parenteral treatments were mainly used in sows and piglets, while oral treatments were more common in nursery and fattening pigs (Moura et al., 2023). Parenteral antimicrobial treatments were often overdosed, while oral treatments were more frequently underdosed (Callens et al., 2012; Timmerman et al., 2006).

There are various indications for treating pigs with antimicrobials, such as gastrointestinal, respiratory or general infections (Jensen et al., 2012; Sarrazin et al., 2019; van Rennings et al., 2015). The most common treatment indication often depends on the pig’s age. Suckling piglets are mainly treated against disorders of the gastro-intestinal, locomotory, respiratory, and nervous system; nursery pigs mainly receive antimicrobial treatment for digestive or locomotory disorders; and the dominant indication for antimicrobial treatment of fattening pigs are gastro-intestinal and respiratory infections. For the sows, urogenital and locomotory infections are the main reason for antimicrobial treatment (Hémonic et al., 2018; Jensen et al., 2012; Moura et al., 2023; Scoppetta et al., 2017).

There is an association between AMU and the phase of pig production. Antimicrobials are most commonly used in younger pigs, especially in nursery pigs (Callens et al., 2012; Sarrazin et al., 2019; Sjölund et al., 2016). Furthermore, there are several peak moments where AMU increases and these peaks probably relate to the following periods of stress in a pig's life: birth and castration (week 1), weaning (week 4) and the start of the fattening phase (week 9) (Figure 2) (Dewulf et al., 2022; Sarrazin et al., 2019). The study of Sarrazin et al. (2019) was performed in nine European countries, i.e. Belgium, Bulgaria, Denmark, France, Germany, Italy, Poland, Spain, and The Netherlands, generally working under the same conditions, where these periods are similar over farms. Previous research showed that strategic prophylactic administration of antimicrobials to entire groups of animals is common practice (Sjölund et al., 2016).

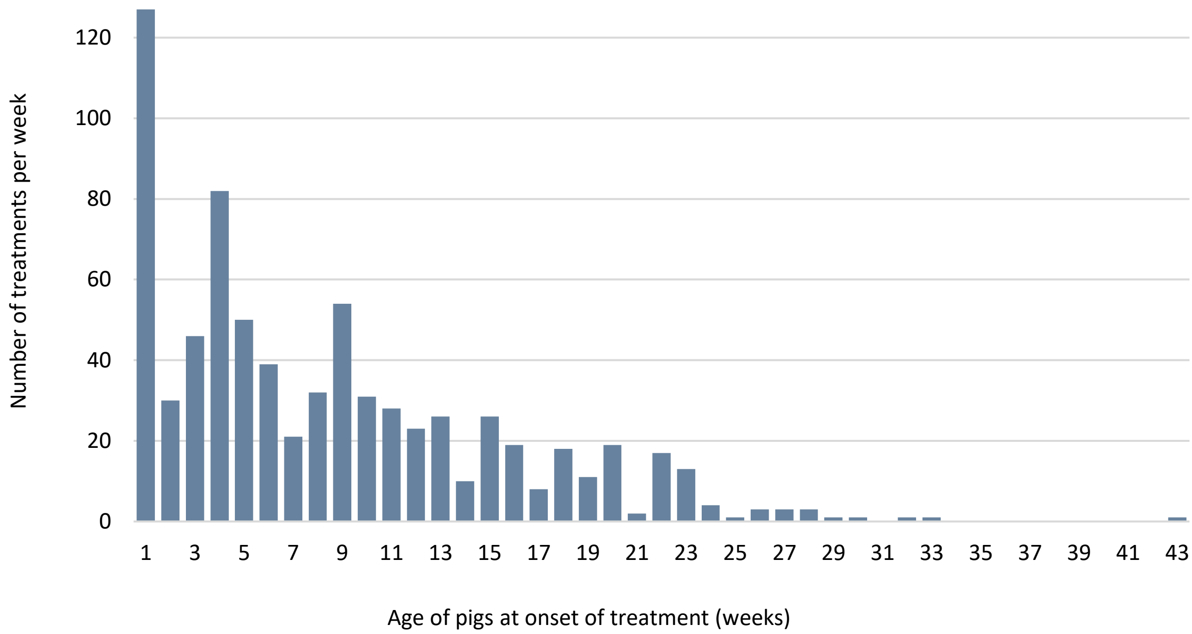

Figure 2. Histogram showing the number of antimicrobial group treatments per week applied to a batch of pigs from birth to slaughter (adapted from Sarrazin et al., 2019).

Several studies have identified factors associated with AMU. These factors can be divided into four main groups, namely: (1) farm characteristics, (2) biosecurity, (3) immunity, and (4) socio-economic factors (Table 1).

Table 1. Overview of the factors associated with antimicrobial use in pig production.

Type of risk factor	Risk factors	References
Farm characteristics	No analysis of production parameters	Arnold et al. (2016)[c], Hirsiger et al. (2015)[b]
	High mortality in fattening pigs	Casal et al. (2007)[a,c], O'Neill (2016)[a]
	Large farms (number of sows)	Backhans et al. (2016)[a], Van der Fels-Klerx et al. (2011)[c,d]
	Small farms (number of fattening pigs)	Hybschmann et al. (2011)[a,c], Vieira et al. (2011)[c]
	Shorter farrowing rhythm	Postma et al. (2016a)[a]
	Visits by herd veterinarian < 2 times per year	Hirsiger et al. (2015)[b]
	No Specific Pathogen Free farm	Hybschmann et al. (2011)[a,c], Sjölund et al. (2015)[a]
	Type of farm (fattening > farrow-to-finish)	Casal et al. (2007)[a,c], Hemme et al. (2018)[b,c], Hybschmann et al. (2011)[a,c], Van der Fels-Klerx et al. (2011)[c,d]
Overall biosecurity	Low overall biosecurity	Collineau et al. (2017a)[a], Laanen et al. (2013)[a], Postma et al. (2016b)[a]
External biosecurity	Low external biosecurity	Postma et al. (2016a)[a]
	Farm staff also working on other farms	Arnold et al. (2016)[c]
	Distance to next pig farm < 500 meter	Arnold et al. (2016)[c]
	Pig-dense area	Hybschmann et al. (2011)[a,c], Van der Fels-Klerx et al. (2011)[d]
	No specific footwear available for visitors	Arnold et al. (2016)[c]
	Poor water quality in farrowing unit	Hirsiger et al. (2015)[b]
Internal biosecurity	Low internal biosecurity	Laanen et al. (2013)[a], Mallioris et al. (2022)[a,d]
	No working routine from healthy to sick animals	Arnold et al. (2016)[c]
	Mixing pigs of different suppliers in the same pen	Arnold et al. (2016)[c]
	Constant supplying of pigs in the compartments	Hirsiger et al. (2015)[b]
Immunity	Nursery pigs	Callens et al. (2012)[a], Sjölund et al. (2016)[a]
	Younger weaning age	Postma et al. (2016a)[a]
	Vaccination of animals	Collineau et al. (2018)[a], O'Neill (2016)[a], Postma et al. (2016a)[a], Stevens et al. (2007)[a,c], Temtem et al. (2016)[a]
Socio-economic factors	Increasing age of the farmer	Backhans et al. (2016)[a]
	Female farmers	Backhans et al. (2016)[a]
	High education of farm staff	Backhans et al. (2016)[a]
	Veterinarian's prescribing behavior	Hybschmann et al. (2011)[a,c], Speksnijder et al. (2015a), Speksnijder et al. (2015b)

The studies were performed on different types of farms: [a] farrow-to-finish; [b] nursery; [c] fattening, [d] farrow-to-wean

The European Surveillance of Veterinary Antimicrobial Consumption (ESVAC) project, initiated by the European Medicines Agency (EMA), collects sales data on veterinary antimicrobials in the European Union. These sales data are expressed in milligrams of active compound per population correction unit (PCU) to correct for the animal population that could be potentially treated in each country. The PCU only includes food-producing animals, including horses and farmed fish, because population data of companion animals are not available for all participating countries. Participation in the project is voluntarily and in the first report of 2010, only 19 EU countries participated, while in the most recent report with data of 2021, 31 countries were already participating (Figure 3). Twenty-five countries continuously provided sales data between 2011 and 2021 and their sales were reduced by 47 % over this period (European Medicines Agency (EMA), 2022). A decrease in AMU is mainly seen in countries with an initial high use. For low-user countries, e.g. Scandinavian countries, it is more difficult to further reduce AMU. These ESVAC data are successful in monitoring the overall trends in AMU in animals in Europe. However, it is difficult to extract the specific evolution in AMU for specific animal species, since many antimicrobials are registered for multiple species. Nonetheless, in several European countries, pig production accounts for a major part of animal production. Therefore, it can be assumed that the observed reductions in antimicrobials are partially due to the reduction of AMU in pig production (Dewulf et al., 2022). When comparing countries, we should consider the composition of the animal population, i.e. the proportion of ruminants vs. monogastric species, since ruminants weigh heavily in the biomass calculation and countries with proportionally more ruminants compared to monogastric animals are favored over countries with relatively more monogastric animals. From the 31 participating countries in 2021, Belgium was the 10th highest user of veterinary antimicrobials. In comparison to neighboring countries, such as Germany, France, and The Netherlands, with a more or less comparable livestock composition, Belgium is definitely not the best student in class and AMU should be further reduced (European Medicines Agency (EMA), 2022).

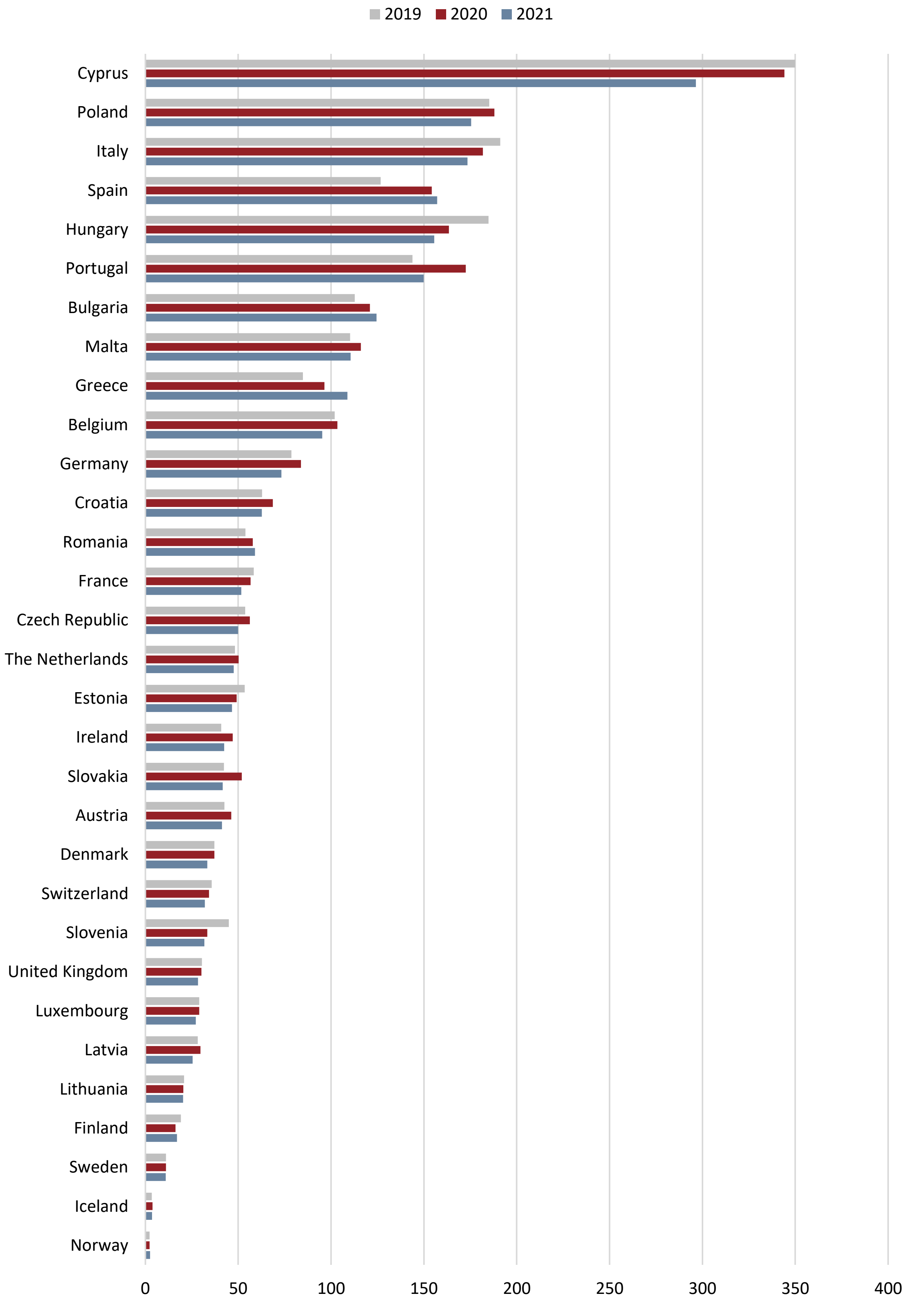

Figure 3. Overview of the total sales of veterinary antimicrobials for food-producing animals in 31 European countries expressed in mg of active compound per PCU (population correction unit) from 2019 to 2021 (adapted from European Medicines Agency (EMA), 2022).

In Belgium, the Belgian Veterinary Surveillance of Antimicrobial Consumption (BelVet-SAC) report yearly describes the AMU of animals. This report relies on sales data (collected at the level of the wholesalers/distributors and the compound feed manufacturers) and data of antimicrobials used (collected at farm-level) in farm animals, as well as in horses and companion animals. The report takes into consideration the amount of antimicrobials that were sold (the numerator) and the biomass (in kg) (the denominator). The biomass is the sum of the amount of meat from beef, pork, poultry and small ruminants plus the number of dairy cattle present in Belgium times 500 kg of metabolic weight per head. Some studies suggest that AMU in pigs is higher compared to other species (Bondt et al., 2013; Merle et al., 2012), but country differences should be considered, as there might be differences in livestock production systems, biosecurity, legislation, and even awareness and skills of veterinarians and farmers (Bondt et al., 2013). In Belgium, veal calves use the most antimicrobials during their lifespan (Figure 4). From 2018 to 2022, a decrease in AMU of 51 %, 31 %, 42 %, and 16 % was established for the suckling piglets, nursery, fattening, and breeding pigs, respectively. Although AMU is decreasing over the years for all animal species, there is still much room for improvement, especially in nursery pigs and veal calves (BelVet-SAC, 2023).

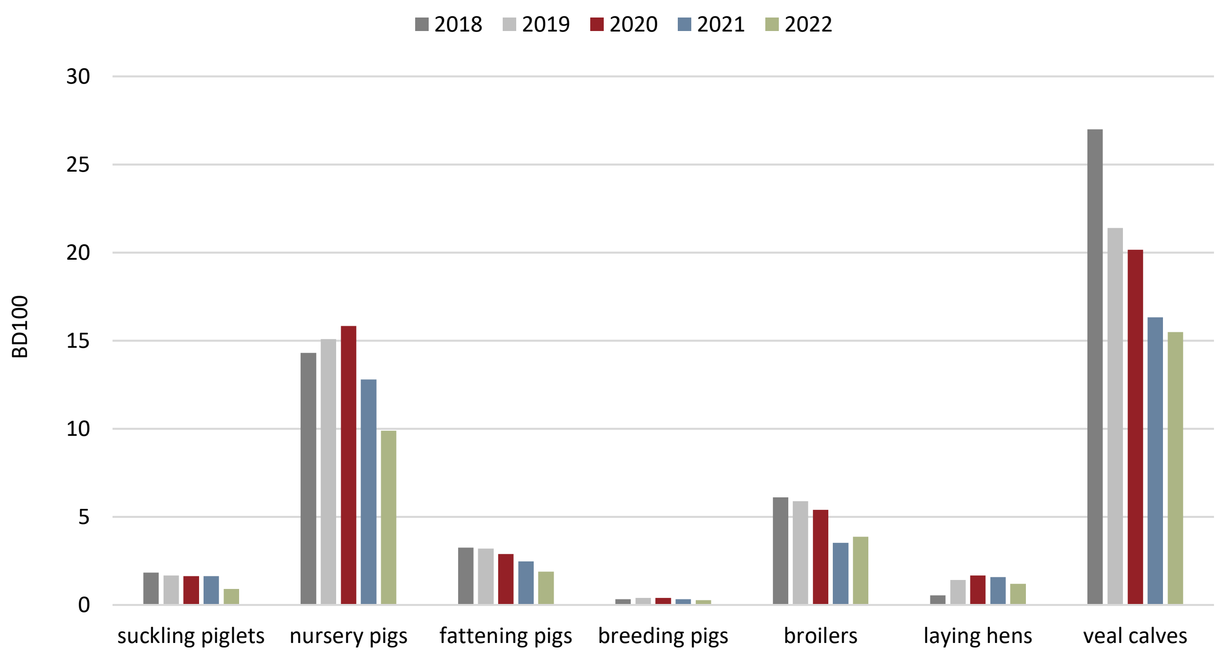

Figure 4. Evolution of the median BD100 (treatment with antimicrobials per 100 days) for different species from 2018 to 2021. Zero-use farms were excluded from the analysis (adapted from BelVet-SAC, 2023).

3. ANTIMICROBIAL RESISTANCE

3.1 ORIGIN, SPREAD, AND PERSISTANCE OF ANTIMICROBIAL RESISTANCE

AMR refers to the ability of bacteria to resist the effect of antimicrobials. AMR is not a new phenomenon; in fact, a study found resistance genes in bacterial DNA from ancient permafrost soil samples dating back thousands of years (Schwarz et al., 2017). AMR can be intrinsic or acquired. Several bacteria are intrinsically resistant to antimicrobials, e.g. *Mycoplasma* spp. are naturally resistant to penicillin and other beta-lactam antibiotics because they lack a cell wall, which is the primary target of these antibiotics. Acquired antimicrobial resistance occurs when bacteria become resistant against antimicrobials that were previously effective in treating infections caused by these bacteria. AMR can be transferred vertically and horizontally: vertical transmission occurs from mother to daughter bacteria through genetic information, while the horizontal spread takes place between different bacteria via specific mechanisms such as transformation, transduction, or conjugation (Chantziaras, 2017; Dewulf, 2018; Schwarz et al., 2017). When penicillin was discovered, Fleming already warned for resistance if penicillin was not used according to the correct dose and duration of treatment (Aminov, 2010).

The widespread use of antimicrobials in human and veterinary medicine, for both farm and companion animals, has accelerated the rate at which bacteria are developing resistance and results in selection pressure for AMR (Chantziaras et al., 2014; Goossens et al., 2005). Different factors influence the impact of AMU on AMR, namely the total AMU, active substance, dose and duration of treatment, administration route, and whether the substance is applied at group vs. individual animal-level (Dewulf, 2018).

Humans and animals, both domestic and wildlife, continuously interact with each other and with the specific environment or ecosystem they live in, this holistic approach is referred to as the One Health concept (Figure 5) (Dewulf, 2018). AMR can be transmitted from animals to humans by three main pathways. This first one is direct contact between animals and humans. Contact between companion animals and owners is possible, but also contact with farm animals by farmers, farm staff, and veterinarians. The second one is spread of AMR via the food chain. This is mainly by consumption of raw or undercooked meat, but AMR can also spread by consuming fruit and vegetables, since animal manure is used as fertilizer on pastures. Finally, interaction with the environment can lead to spread of AMR. If environmental bacteria come into contact with antimicrobial residues present in manure and water, AMR can spread via the environment. Once there is selection and spread of AMR, it is difficult to get rid of, resulting in persistence of AMR (Dewulf, 2018).

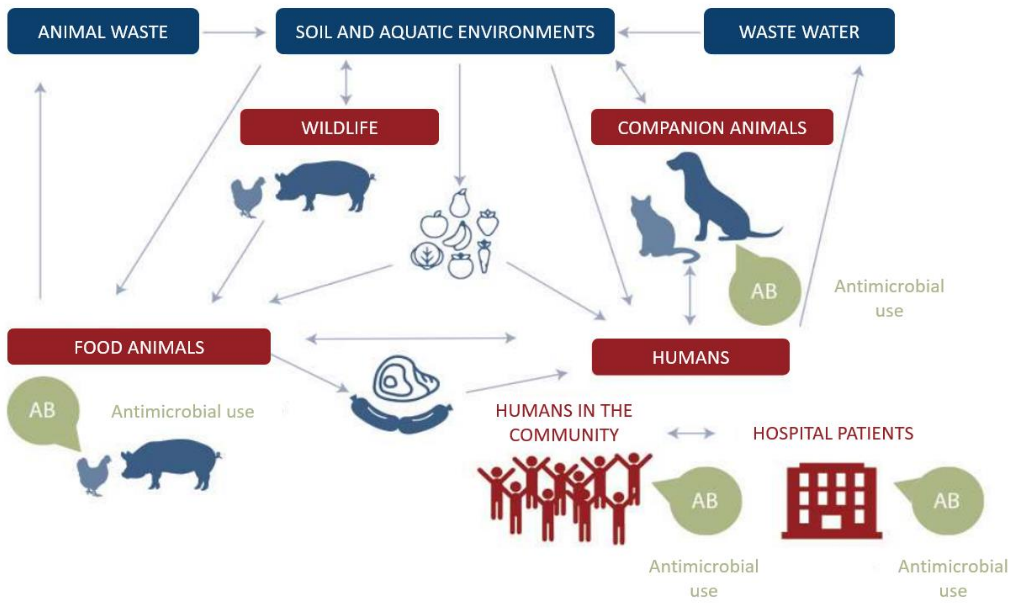

Figure 5. Various routes for the exchange of resistant bacteria between humans, animals, the environment, both through direct and indirect contact (Dewulf, 2018).

3.2 CONSEQUENCES OF ANTIMICROBIAL RESISTANCE

When bacteria develop AMR, antimicrobials become ineffective in killing or controlling the growth, making it difficult to treat infections caused by these bacteria. This can lead to therapy failure, increased disease, and mortality. Several resistant bacteria can cause disease in pigs and The European Food Safety Authority (EFSA) identified *Escherichia coli* and *Brachyspira hyodysenteriae*, both causing gastro-intestinal infections in pigs, as the most relevant antimicrobial resistant bacteria in the EU (EFSA Panel of Animal Health and Welfare, 2021).

AMR is a growing public health concern. To address this problem, a One Health approach is required, where collaboration between veterinary and human health care professionals is important. Responsible use of antimicrobials, increased infection prevention, and alternative therapies can be helpful to tackle AMR.

Since 2011, all EU Member States should monitor and report AMR in food-producing animals. The monitoring is performed for different bacteria, i.e. indicator commensal *E. coli*, and zoonotic *Salmonella* and *Campylobacter* spp.; and for different food-producing animals and foods. For pigs, fattening pigs and fresh pig meat should be monitored (European Commission, 2020). The goal of this program is to monitor the sensitivity of *E. coli* against antimicrobial classes that are important for animal and public health. Multi-resistant *E. coli* strains refer to strains that are resistant to at least 3 out of 12 tested antimicrobial classes. In fattening pigs in Belgium, the multi-resistant *E. coli* have decreased from 47.8 % to 28.8 % and the prevalence of sensitive *E. coli* strains have increased from 28.8 % to 41.8 % in 2014 and 2021, respectively

(Federaal Agentschap voor de Veiligheid van de Voedselketen (FAVV), 2022). In 2022, the multi-resistant *E. coli* increased, while the prevalence of sensitive *E. coli* strains decreased. Even though the results from 2022 are less promising compared to 2021, there is still an overall improvement in sensitivity when comparing to 2011 (Federaal Agentschap voor de Veiligheid van de Voedselketen (FAVV), 2023). Although a reduction of AMR is seen in the past years, the level of AMR is still high and action should be taken to further decrease AMU and subsequently AMR.

4. ANTIMICROBIAL STEWARDSHIP

AMR is a significant global health concern and the emergence and spread of AMR is driven by the over- and misuse of antimicrobials in both human and veterinary medicine. AMR highlights the urgent need to reduce AMU. To do so, implementing prudent and responsible AMU is crucial. Antimicrobial stewardship refers to strategies and interventions aimed at optimizing the use of antimicrobials to minimize the emergence of AMR and to preserve the effectiveness of antimicrobials for the future.

4.1 ANTIMICROBIAL STEWARDSHIP IN EUROPE

Different organizations categorized antimicrobials according to their importance. The World Health Organization (WHO) ranked antimicrobials according to their importance in human medicine and classified them in three categories: (1) important, (2) highly important, and (3) critically important. The criteria used for assigning antimicrobials to the critically important category were: (1) sole therapy or one of few alternatives to treat serious human disease, and (2) antimicrobial used to treat diseases caused by organisms that may be transmitted via non-human sources or diseases caused by organisms that may acquire resistance genes from non-human sources. Important antimicrobials meet neither criterium 1 nor 2, highly important antimicrobials meet criterium 1 or 2, and critically important antimicrobials (CIA) meet both criteria 1 and 2 (Collignon et al., 2009; World Health Organization (WHO), 2005).

Also the World Organization for Animal Health (WOAH) categorized antimicrobials, but based on their importance in veterinary medicine. The categories are similar to those of human medicine, namely: (1) veterinary important antimicrobials, (2) veterinary highly important antimicrobials, and (3) veterinary critically important antimicrobials. The categorization was based on two criteria: (1) response rate to the questionnaire to WOAH Delegates of all Member Countries regarding Veterinary Critically Important Antimicrobials, and (2) treatment of serious animal disease and availability of alternative antimicrobials (World Organization for Animal Health (WOAH), 2007).

The EMA combined the previous two categories into a categorization of antimicrobials for use in animals. In this list, antimicrobials are categorized based on the potential consequences of AMR to public health, but taking into account the need for their use in veterinary medicine.

The list includes four categories: (1) Category A (avoid), (2) Category B (restrict), (3) Category C (caution), and (4) Category D (prudence) (European Medicines Agency (EMA), 2020).

- Category A (avoid) antimicrobials are not authorized as veterinary medicines in the EU and they should not be used in food-producing animals. Under exceptional circumstances, they can be given to companion animals.
- Category B (restrict) antimicrobials are critically important in human medicine and use in animals should be restricted to mitigate the risk to public health. They should only be considered when there are no antimicrobials in category C or D that could be clinically effective and the use should be based on antimicrobial susceptibility testing.
- Category C (caution) antimicrobials have alternatives in human medicine and for some veterinary indications there are no alternatives from category D. They should only be considered when there are no antimicrobials in category D that could be clinically effective.
- Category D (prudence) antimicrobials should be used as first line treatments, whenever possible. As always, these antimicrobials should be used prudently and only when medically needed.

Furthermore, EMA recommends that unnecessary long treatment periods and under-dosing should be avoided. Group treatments should be restricted to situations where individual treatment is not feasible. Furthermore, the administration route should be taken into consideration when prescribing antimicrobials. The following list ranks the administration routes and formulation types from lowest to highest estimated impact on AMR.

- Local individual treatment (e.g. udder injector, eye or ear drops)
- Parenteral individual treatment (intravenously, intramuscularly, subcutaneously)
- Oral individual treatment (i.e. tablets, oral bolus)
- Injectable group medication (metaphylaxis), only if appropriately justified
- Oral group medication via drinking water/milk replacer (metaphylaxis), only if appropriately justified
- Oral group medication via feed or premixes (metaphylaxis), only if appropriately justified

Collecting objective data is crucial for the development of reduction strategies. Therefore, quantifying farm-level AMU is important. However, there is no standardized method in Europe for data collection, analysis methodology, or benchmarking strategies, leading to non-comparable outcomes in different studies. In the European AACTING project (short for "Network on quantification of veterinary Antimicrobial use at herd level and Analysis, CommunicaTion and benchmarkING to improve responsible use"), guidelines were developed with practical advice for setting up data collection systems at farm-level (AACTING consortium, 2019). Furthermore, a review of existing AMU monitoring systems (n = 38) in different countries (n = 16) for different species (n = 13) was made publicly available (Sanders et al., 2020).

Currently, the ESVAC-project already shows sales data of veterinary antimicrobials of 31 countries. However, participation in this project is voluntarily and there is no distinction between different animal species. The new EU regulation on Veterinary Medicinal Products takes this a step further; and from 2023 onwards, a stepwise approach to monitor AMU in animals on species-level will be mandatory for all EU Member States (European Council Regulation, 2018).

- 2023: pigs, broilers, laying hens, turkeys, veal calves, dairy cattle, beef cattle
- 2026: all food-producing animals, including other poultry (ducks, geese), small ruminants (sheep, goat), fish, rabbits, and horses
- 2029: all animals which are bred or kept, including dogs, cats, and fur animals

Also in legislation, efforts have been made to raise awareness on AMR and providing alternatives for AMU. Regulation 2016/429 on transmissible animal diseases, also referred to as the new Animal Health Law, focuses on disease prevention, including biosecurity (European Council Regulation, 2016). Regulation 2019/6 of the European Parliament and the Council of 11 December 2018 on Veterinary Medicinal Products emphasizes the One Health approach and the prudent use of antimicrobials (European Council Regulation, 2018).

4.2 BELGIAN EFFORTS TO REDUCE ANTIMICROBIAL USE

4.2.1 AMCRA

In 2012, the knowledge center on AntiMiCrobial use and Resistance in Animals in Belgium (AMCRA) was founded as a non-profit organization. The main goal of AMCRA is a reduction of AMU by analysis, communication, and sensitization for both farm (pigs, poultry, and cattle) and companion animals, with the goal to safeguard both human and animal health.

4.2.1.1 TASK 1: REDUCTION PLANS AND TARGETS FOR AMU IN BELGIUM

The first task of AMCRA is generating reduction plans and targets for AMU in Belgium. The first reduction plan was called ‘Vision 2020’. This plan contained three goals with 2011 as the reference year (AMCRA, 2016).

1. 50 % reduction of AMU by 2020
2. 75 % reduction of the CIA by 2020
3. 50 % reduction of antibiotic medicated feed by 2017

Unfortunately, not all goals were achieved by 2020. The total AMU and the CIA were reduced by 40.2 % and 70.1 %, respectively. These are significant reductions; however, it was insufficient to meet the predetermined goals. The antibiotic medicated feed decreased with 70.4 % (Federaal Agentschap voor de Veiligheid van de Voedselketen (FAVV), 2021a).

In 2019, AMCRA designed a new reduction plan called 'Vision 2024', where four strategic objectives regarding AMU and AMR in animals in Belgium between 2021 and 2024 are described, with 2011 being again the reference year. The ultimate goal is to reach a minimal AMU in all animal species and by all veterinarians. In order to achieve these four objectives, nine action points were suggested in 'Vision 2024' (Figure 6) (AMCRA, 2019).

1. Species-specific thresholds at farm-level and no more than 1 % very high AM users by 2024
2. Total AMU evolves towards the median use in Europe by 2024 (corresponding to a 65 % reduction compared to 2011)
3. Maximum use of 1 mg colistin/kg biomass by 2024
4. A 75 % reduction of antibiotic medicated feed by 2024

Figure 6. Nine action points from AMCRA's 'Vision 2024' (AMCRA, 2019).

Preliminary results show that total AMU decreased by 58.2 % (target 65 %), the amount of colistin was 0.5 mg/kg biomass (target 1 mg/kg), the reduction of antibiotic medicated feed was 83.5 % (target 75 %), and the amount of CIA were reduced with 82.7 % (target 75 %) from 2011 until 2022. In pigs, there were 4,5 % very high AM users (target 1 %). Almost all targets were already reached in 2022, except for the decrease in total AMU and the percentage of very high AM users (Federaal Agentschap voor de Veiligheid van de Voedselketen (FAVV), 2023).

4.2.1.2 TASK 2: ANTIMICROBIAL STEWARDSHIP

The second main task of AMCRA is to promote antimicrobial stewardship in the field of veterinary medicine. Prudent AMU is important; therefore, AMCRA designed guidelines for AMU. By the means of a formulary, a stepwise decision of antimicrobials by the veterinarian is possible when a treatment with antimicrobials is justified. The formulary gives an overview of antimicrobials for different indications. Antimicrobials are categorized in first, second, and third choice products. This categorization is based on scientific data regarding antibacterial susceptibility, pharmacokinetics and -dynamics, and clinical efficacity. First choice antimicrobials are preferred over second choice and second choice are preferred over third choice antimicrobials. Antimicrobials are also divided into groups according to color codes based on the importance of the molecule in public and veterinary health (Table 2). Within one particular choice (first, second, or third choice), antimicrobials with different color codes may be classified. Antimicrobial susceptibility testing may indicate multiple treatment options with different color codes. Preference should then be given to the antimicrobial of least public health concern, i.e. first yellow, then orange, and red as the last option (AMCRA, 2022b).

Table 2. Terms of use for the AMCRA color codes.

	Additional laboratory analysis	**Antimicrobial susceptibility test**
Yellow	recommended	recommended
Orange	mandatory	recommended
Red	mandatory	mandatory

The veterinarian should choose a product that is registered for the animal species and indication. Furthermore, the administration route of antimicrobials plays a role in the selection pressure: individual treatment is always preferred over group treatment and local or parenteral treatments are preferred over oral treatments. These guidelines are regularly revised and updated. They are also used as a reference by veterinary students from the universities of Ghent and Liege. In addition, AMCRA has also drawn up guidelines for sampling and diagnostics. AMCRA also formulates recommendations in order to answer to specific scientific or technical questions. In order to reach the farmers and veterinarians, AMCRA uses different channels for sensitization and communication, namely digital newsletters, press releases, articles in magazines, presentations and participation at events and fairs, and social media.

4.2.1.3 TASK 3: QUANTIFICATION OF AMU

The third main task of AMCRA is the quantification of AMU. In Belgium, mandatory registration of veterinary antimicrobials in the national data collection system, i.e. Sanitel-Med, evolved over the years (Table 3). Legislation requires the registration of antimicrobials directly in Sanitel-Med for pigs, broilers, laying hens, and veal calves since 2017; and for all cattle and poultry (chicken and turkey) since 2023

(Royal Decree, 2023). In addition, various quality labels impose a mandatory affiliation with AB Register, which is linked to Sanitel-Med. Besides the mandatory affiliation with AB Register, there is also the possibility of joining voluntarily (AB Register, 2023). After registration of the antimicrobials in the database, the data analysis unit of AMCRA, which was founded in 2014, performs the analysis. Subsequently, benchmarking reports are sent to farmers and their herd veterinarian.

Table 3. Timeline of mandatory veterinary antimicrobial registration in different animal species in Belgium, imposed by legislation or quality labels (AB Register, 2023; Royal Decree, 2023).

Year	Species	Quality label/legislation	Database
2014	Pigs	BePork	AB Register
2017	Pigs, broilers, laying hens, veal calves	legislation	Sanitel-Med
2017	Poultry	Belplume	AB Register
2018	Dairy cattle	IKM Vlaanderen	AB Register
2022	Beef cattle	Belbeef	AB Register
2023	Cattle, poultry (chicken and turkey)	legislation	Sanitel-Med

AMCRA calculates the BD100 (‘BehandelDagen met antibiotica per 100 dagen’), which is a standardized way to quantify AMU. The BD100 is the number of treatment days with antimicrobials in 100 days or the percentage of treatment days with antimicrobials; and it is calculated for the suckling piglets, nursery, fattening, and breeding pigs.

$$\text{BD100} = \frac{\text{amount of antibiotics administered (mg)}}{\text{DDDa}_{\text{bel}} * \text{kg animal 'at risk'} * \text{number of days 'at risk'}} * \text{LA}_{\text{bel}} * 100$$

The numerator of the formula consists of the amount of antimicrobials administered (expressed in milligram) and the long-acting factor (LA_{bel}), which corrects for products with an active duration longer than 24 hours (AMCRA, 2022d). In the denominator, the Belgian defined daily dose for animals is used ($DDDA_{bel}$) (AMCRA, 2022c). These values are defined based on the information in the summary of product characteristics (SPC) for each antimicrobial. The total weight of animals at risk for treatment is the average number of animals on the farm multiplied by a standardized weight at treatment. The average number of animals within each animal category is determined for each farm. For the suckling piglets, this is determined by multiplying the number of sows in a herd by 30, i.e. the average number of weaned piglets per sow per year, divided by 12, i.e. number of months per year. The standardized weight of the pigs at treatment is defined as 4 kg, 12 kg, 50 kg and 220 kg for the different animal categories, respectively. The number of days animals are at risk to receive treatment is also included, namely 30.42, i.e. the average length of a month. Regardless of the number of days at risk, the AMU is always converted to 100 days.

AMU is then compared to benchmarking values which are determined for the different animal categories. Before, the benchmarking values were determined per animal category based on the frequency distribution of the BD100 values in each category. The 50th and 90th percentiles of the frequency distribution were used as alert and action values, respectively, meaning that the alert value was determined by the 50 % lowest users and the action value was determined by the 10 % highest users, resulting in dynamical benchmarking values. However, these benchmarking values were later adapted to stricter fixed values. The alert and action values of the BD100 define three user categories: (1) low-user, (2) alert-user, and (3) high-user farms, when the BD100 is below the alert value, between the alert and the action value, and when the BD100 exceeds the action value, respectively (Figure 7). Low-user farms are considered to be in the safe zone with a non-concerning AMU, but these farmers should strive to keep their AMU low. Alert-user farms should pay extra attention to their AMU and they should try to reduce it. High-user farms should immediately take action to reduce AMU. Farms with a long-term or repeated high AMU must be assisted by an external coach to reduce their AMU. These specialized external coaches received a training by the government and will support farmers and their veterinarian.

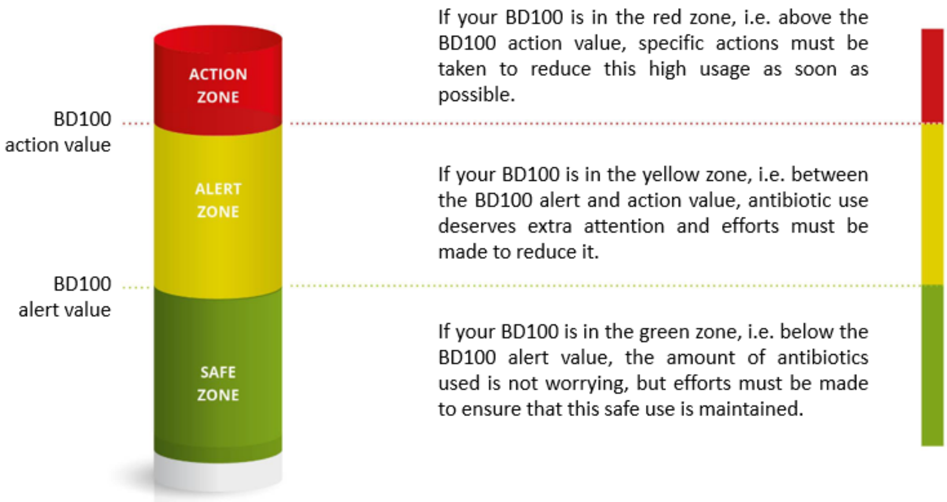

Figure 7. The Belgian benchmarking system is defined by two thresholds: the alert and action value (AMCRA, 2022a).

The benchmarking reports are sent to the farmers two or four times per year, depending on whether they are affiliated to Sanitel-Med or AB Register, respectively. Since 2019, AMCRA provides benchmarking reports for veterinarians and veterinary practices, since veterinarians also play an important role in AMU on farms.

4.2.2 GOVERNMENT

In 2016, the first covenant in the fight against AMR on prudent use of antimicrobials in animals was established between the Belgian Federal Government and various partners involved in animal production. This covenant defined ambitious targets to reduce AMU in animals from 2016 to 2020. To continue this collaboration, a second covenant for the period 2021 to 2024 was signed with the various partners, together with the government, committing to achieving new reduction targets while maintaining low use for the critically important antimicrobials (Federaal Agentschap voor de Veiligheid van de Voedselketen (FAVV), 2021b).

Each EU Member State is obliged to develop a One Health action plan to combat AMR (European Commission, 2017b; World Health Organization (WHO), 2015). In Belgium, this plan contains ten strategic directions. These ten strategic directions were further broken down into operational objectives (*n* = 76) and actions (*n* = 230) (Figure 8) (Federale Overheidsdienst Volksgezondheid Veiligheid van de Voedselketen en Leefmilieu (FOD VVVL), 2020).

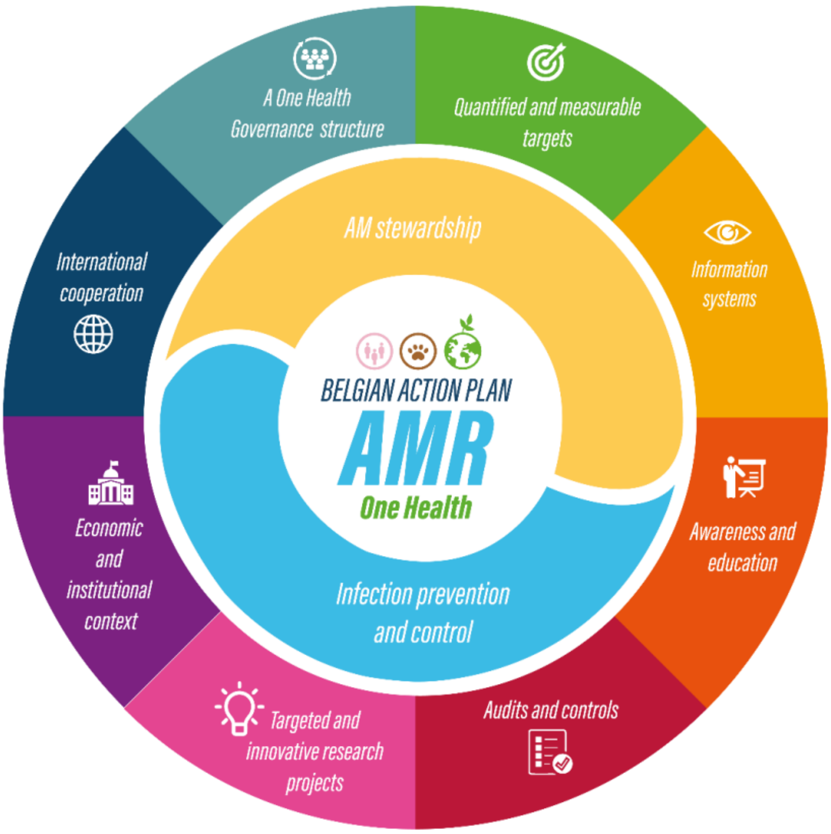

Figure 8. Ten strategic directions for combatting AMR in the Belgian One Health national action plan (Federale Overheidsdienst Volksgezondheid Veiligheid van de Voedselketen en Leefmilieu (FOD VVVL), 2020).

4.2.3 QUALITY LABELS

Different Belgian quality labels impose, in addition to national legislation, extra guarantees to improve animal welfare, sustainability, and animal health. Quite often, important aspects of these labels' guidelines focus on AMU. Pig producers can voluntarily participate in these labels. Table 4 shows an overview of the criteria regarding antimicrobials in two important Belgian quality labels, namely BePork and Beter Leven. BePork is a Belgian quality label that is equivalent to the German label QS ('Qualitätssicherung'), guaranteeing export to Germany. This label guarantees local production 'from farm to fork' through the entire production chain, including feeding companies, pig farms, transporting companies, slaughterhouses, etc. Currently, 90 % of the Belgian pig farmers produce according to this label (BePork, 2022). Since most Belgian pig farmers participate in BePork, other quality labels, such as 'Colruyt' and 'Duroc d'Olives', require affiliation with BePork as well. Furthermore, additional measures, e.g. in terms of animal welfare and public health, are requested. However, the specification of these labels are not publicly available online. Beter Leven is a Dutch quality label that was introduced in Belgium in 2017. A three-star-level indicates the adjustments that have been made for animal welfare; and three stars correspond to organic production. For all three levels, the criteria regarding antimicrobials are the same (Beter Leven, 2022).

Table 4. Overview of criteria regarding antimicrobials in two Belgian quality labels.

	BePork	**Beter Leven**
Registration of AMU in national database (AB Register)	x	x
Obligatory consultation of periodic benchmarking reports	x	–
Measures when AMU exceeds action value	x	x
External expert in case of long-term high AMU	x	x
Farm-specific herd health plan	x	x
Farm-specific treatment protocol	(x)	x
Special measures regarding CIA (e.g. AM susceptibility test)	x	x
No topdressing in feed trough	x	x
No adding of AM on the farm itself by 'mills'	x	x
No medicated feed	–	x

x: yes; (x): yes, in some cases; –: no

4.3 RAISED WITHOUT ANTIBIOTICS

A reduction of antibiotic-resistant bacterial isolates in pig production can be obtained by prudent AMU, but also by the restriction of antimicrobials. An example of the latter is the Raised Without Antibiotics (RWA) concept in which pigs are raised without the use of any antibiotics from birth until slaughter. The concept is recognized in only a few countries. According to the Danish Integrated Antimicrobial Resistance

Monitoring and Research Program (DANMAP), 51 pig farms in Denmark raised pigs without antibiotics in 2018 (DANMAP, 2018). Two studies, each investigating two RWA sow farms, further examined RWA production in Danish pig farms (Baekbo, 2017; Lynegaard et al., 2021). In the United States, RWA is an independent certification that covers all animal source foods including meat, poultry, seafood, fish, dairy, and eggs. It is certified by the National Sanitation Foundation (National Sanitation Foundation, 2023; Singer et al., 2019). In The Netherlands, there is also an antibiotic-free concept called 'Antibioticavrij Leven Garantie' (Duurzaam Varkensvlees, 2023); and also in Poland (Cybulski et al., 2021), and Canada (Alvarado et al., 2022) the RWA concept is known. However, on a larger scale, it is unclear what the characteristics of RWA farms are, as well as which differences exist in comparison to conventional pig farms. Furthermore, the specific inclusion criteria for RWA production are not well specified in literature and the implementation of RWA in a larger number of farms with varying management and housing conditions requires further investigation.

5. ALTERNATIVES FOR ANTIMICROBIAL USE

Besides antimicrobial stewardship, alternatives for antimicrobials could also help reducing AMU. This paragraph will focus on different alternatives, i.e. accurate diagnostics, biosecurity, vaccination, feed (additives), stable climate and housing, and coaching. Obviously, one alternative method cannot completely replace the use of antimicrobials. The more alternative (and preventive) measures that can be implemented, the better.

5.1 ACCURATE DIAGNOSTICS

Even though it is not really an alternative for antimicrobials, it is important to start this chapter with the importance of accurate diagnostics. Diagnostics play a crucial role in reducing AMU in pig production. Regular diagnostic testing was in the top five of alternatives for AMU with the highest perceived return on investment (ROI) (Postma et al., 2015). Accurate diagnostics are important to correctly identify the cause of disease in order to avoid unnecessary AMU. Accurate disease identification allows for targeted and appropriate use of antimicrobials. By identifying the specific pathogens responsible for an infection, veterinarians can prescribe the right antimicrobials that are effective against those pathogens, avoiding the unnecessary use of broad-spectrum antibiotics.

Frequent diagnostics can determine the herd health status. Regular screening and monitoring can help detect diseases at an early stage. This allows for proactive measures to be taken. Herd-specific action plans can be developed and preventive measures, such as vaccination, can be taken to avoid specific pathogens or problems (Postma et al., 2017).

5.2 BIOSECURITY

5.2.1 INTRODUCTION TO BIOSECURITY

The first citation of the term biosecurity in PubMed was only in 1987. From then on, the topic was described more frequently, with five (in the 1990s), 127 (from 2000 to 2010), and 680 citations (from 2011 until 2020) (Renault et al., 2022). In 1999, Amass & Clark recognized the importance of biosecurity for animal health. At that time, there was no scientific evidence for biosecurity, but they already suggested that registration of visitors, pig movements, etc. needed to be collected, to recognize and address specific violations against biosecurity for reduction of disease.

Biosecurity should be the basis of any infectious disease control program, because it is a proactive approach to prevent disease spread and to protect animal health. By implementing biosecurity measures, disease transmission can be minimized, resulting in less need for curative antimicrobial treatments (Figure 9).

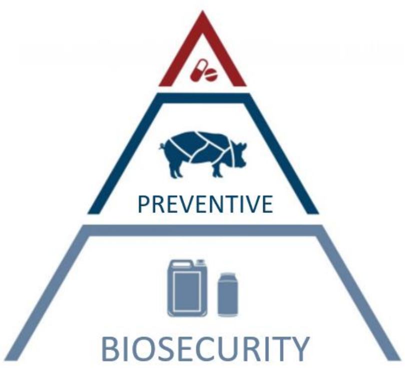

Figure 9. Biosecurity should be the basis of any disease control program (Dewulf, 2018).

Biosecurity measures aim to limit or even fully prevent the transmission of pathogens. All measures that aim to reduce the risk of pathogen introduction on a farm are grouped as external biosecurity measures, while those that aim to reduce the spread of pathogens within a farm are grouped as internal biosecurity measures (Figure 10) (Dewulf et al., 2018).

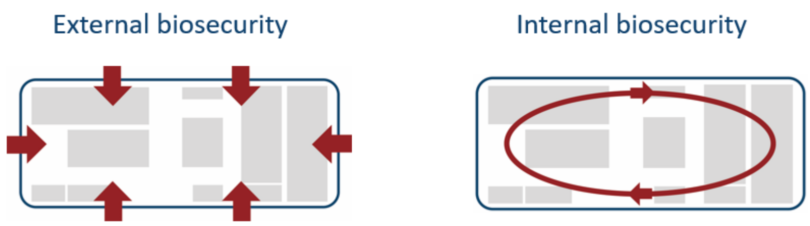

Figure 10. Graphical representation of the external and internal biosecurity (Dewulf, 2018).

Some general principles regarding biosecurity apply for different animal species (Dewulf & Van Immerseel, 2018). These will be discussed more in detail in the next paragraphs.

5.2.1.1 SEPARATION OF HIGH- AND LOW-RISK ANIMALS AND ENVIRONMENTS

Separating high- and low-risk animals and environments is crucial to avoid disease transmission. Preventing direct contact between infectious and susceptible animals is important, but also indirect transmission, e.g. by people, vermin, or equipment, should be avoided. If this contact cannot be ruled out, some precautionary measures should be taken, e.g. quarantine measures, farm-specific clothing, etc. (Dewulf & Van Immerseel, 2018).

Also the separation of clean and dirty areas on the farm is important. The clean area includes the barns, offices, and areas for internal movements on the farm, while the dirty area is for external transport, e.g. feed company, animal transport, or rendering company. As transport vehicles have contact with other farms or slaughterhouses, they have a high risk of introducing pathogens on a farm. Therefore, all external transport vehicles should remain in the dirty area (Alarcón et al., 2021; Dewulf, 2018).

5.2.1.2 REDUCTION OF THE GENERAL INFECTION PRESSURE

Even on farms with the best biosecurity measures, it’s impossible to keep the environment sterile. The overall aim of biosecurity measures is to reduce the general infection pressure to an acceptable level where the immune system of the pigs can take over (Dewulf & Van Immerseel, 2018).

5.2.1.3 LARGER ANIMAL GROUPS POSE HIGHER RISKS

Larger herds pose higher risks and require more biosecurity measures for several reasons. On larger herds there are more animals that are susceptible for infections and in addition, more animals can maintain the infection cycle, increasing the infection pressure. Larger herds also have more contact with the outer world, e.g. by purchasing breeding gilts and more animal transport, which also include the risk of pathogen introduction. Furthermore, in intensive livestock production, high-productive animals tend to be more vulnerable and the consequences of pathogen introduction could be more severe (Dewulf & Van Immerseel, 2018).

5.2.1.4 NOT ALL TRANSMISSION ROUTES ARE OF EQUAL IMPORTANCE

There are different routes of disease transmission and not all routes include the same risk. Ranking these transmission routes according to their relevance may depend on several factors, including the specific pathogens, their survival rate in the environment, and their ability to infect animals. Also the frequency contributes to the risk of the transmission routes: a less important transmission route could become important if this transmission route occurs multiple times. The combined risk (chance of transmission x frequency) can be calculated with a formula, where P is the combined risk (probability), p is the risk of disease transmission per event, and *n* is the number of events.

$$P = 1 - (1 - p)^n$$

Boklund (2008) categorized the routes of disease transmission from low to high risk (Figure 11). This figure implies that not all biosecurity measures contribute equally to the prevention of infectious diseases. This dissertation covers the two routes with the highest risk, i.e. live animals and persons, and these transmission routes will be discussed more in detail further (see 5.2.3 and 5.2.4).

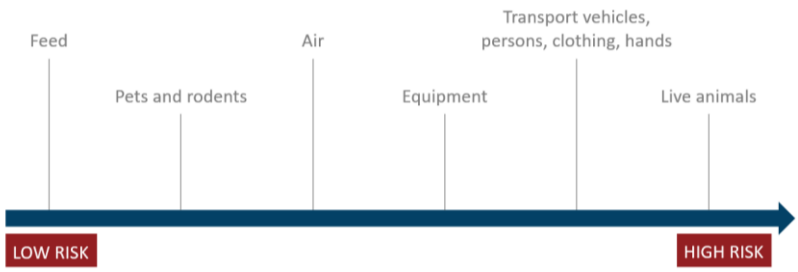

Figure 11. Routes of disease transmission from low to high risk (adapted from Boklund, 2008).

5.2.2 BIOCHECK.UGENT

If you want to improve biosecurity, the first step is to have a thorough understanding of the current status. Therefore, in 2008, the Veterinary Epidemiology Unit of Ghent University developed a risk-based biosecurity quantification tool, Biocheck.UGent, which allows for objectively quantifying the farm's biosecurity status. It is available for pigs, poultry, and cattle. For pigs, the questionnaire consists of 109 questions. Depending on the importance of a particular biosecurity measure, the score of a question is multiplied by a weight factor. In the Biocheck.UGent pig survey, there are six categories of external biosecurity, namely: (1) purchase of breeding pigs, piglets, and semen; (2) transport of animals, removal of carcasses and manure; (3) supply of feed, water, and equipment; (4) visitors and farm staff; (5) vermin and bird control; and (6) location of the farm. There are also six categories of internal biosecurity: (1) disease management; (2) farrowing and suckling period; (3) nursery unit; (4) fattening unit; (5) measures between compartments, working lines, and use of equipment; and (6) cleaning and disinfection. Each category is scored between 0 and 100 %. Zero means a total absence of the described biosecurity measures, while 100 means full application of the described measures (Biocheck.UGent, 2023).

Biocheck.UGent's internal biosecurity classification is based on the different animal compartments (Biocheck.UGent, 2023). However, other classifications can be made based on the applied biosecurity principles, for example (1) disease management; (2) all-in/all-out; (3) stocking density; (4) compartmentalization and working lines; and (5) cleaning and disinfection (Dewulf & Van Immerseel, 2018).

Biocheck.UGent has different services to improve knowledge on biosecurity. Biocheck.UGent offers e-learning modules for farmers and veterinarians with short online lectures to improve knowledge on biosecurity. Biocheck.UGent also provides courses, which are actually train the trainer programs. This way, knowledge on biosecurity can be shared to farmers by (herd) veterinarians.

5.2.3 INTRODUCTION PROCEDURES OF BREEDING GILTS

The breeding population is very important in pig herds, for productivity, health, and profitability (Patterson & Foxcroft, 2019). Replacement of breeding animals can be accomplished by own rearing of breeding gilts or by purchasing them from specialized breeding farms. In Europe, both methods are used about equally (Bernaerdt et al., 2021; Caekebeke et al., 2020; Chantziaras et al., 2018). In the North American swine industry it is common to house breeding gilts in a specialized gilt development unit (GDU), allowing proper selection and management of breeding gilts. GDUs are used to raise gilts and to gradually adapt them to the health status of the sow herd (Garza-Moreno et al., 2018; Williams et al., 2005). Breeding gilts enter these facilities at a young age, allowing sufficient time for acclimation through vaccination or exposure to farm-specific pathogens (Zimmermann et al., 2019).

Purchasing of breeding gilts might lead to faster improvement of genetic potential (Pritchard et al., 2005). However, direct contact between live animals is considered to be the most important and effective pathogen transmission route, thus purchasing gilts includes the risk of pathogen introduction in a farm (Boklund, 2008; Dewulf et al., 2018). The yearly replacement rate on sow farms is around 40 – 45 % (Driessen & Van Thielen, 2012). This means that new breeding stock is frequently introduced in sow farms. Purchased animals should first be housed in a quarantine unit, which aims (1) to reduce the risk of pathogen introduction into the farm, and (2) to facilitate the introduction of the animals into the herd by means of acclimation. During the quarantine period, pigs can be observed for the presence of clinical signs and be tested for the presence of pathogens. Acclimation practices such as vaccination against several pathogens and exposure to live animals (e.g. pigs or sows before culling), can protect newly purchased animals against pathogens circulating on the farm (Garza-Moreno et al., 2017; Pritchard et al., 2005).

The introduction of breeding gilts into a pig farm should be done with careful consideration of various factors, including biosecurity measures. If animals need to be purchased, the farmer should pay attention to the following items to minimize the risk of pathogen introduction: the pigs should always originate from the same farm, the health status of the origin farm should be higher than or equal to the farm, and strict hygiene measures should be implemented for the transport vehicle. Once the newly purchased pigs arrive on the farm, there are some requirements for the building and the management of the quarantine unit. A quarantine unit should be present and ideally, it should be completely isolated from the other animals, with a separate entrance and a separate hygiene lock. The all-in/all-out principle should be practiced, so that new gilts can only arrive when the quarantine unit is completely empty. Finally, the duration of the quarantine period should be a minimum of 28 days (Dewulf et al., 2018). By implementing these biosecurity measures, the risk of introducing diseases through breeding gilts can be minimized, helping to maintain the health and productivity of the pig farm.

Purchasing gilts from specific pathogen free (SPF) farms could be useful to keep the farms free of specific diseases. The number of SPF farms in Belgium is not known. However, the Danish SPF system was introduced in 1971 and currently 2500 Danish herds have the SPF status. There are seven pathogens in the Danish SPF system: *Mycoplasma hyopneumoniae*, *Actinobacillus pleuropneumoniae*, *Pasteurella multocida*, *Brachyspira hyodysenteriae*, porcine reproductive and respiratory syndrome virus (PRRSV), *Sarcoptes scabiei* var. suis, and *Haematopinus suis*. Denmark has 220 nucleus and multiplier herds, which are all SPF, indicating that 100 % of all purchased breeding stock is SPF. Furthermore, there are also 2300 SPF sow and finisher herds, with 80 %, 73 %, and 34 % of all sows, nursery, and fattening pigs, respectively, being SPF (T.S. Hansen, personal communication).

Legislation on the introduction of breeding gilts is covered by the Animal Health Law and focuses on prevention of disease spread between countries by health requirements, veterinary inspections, and quarantine procedures. If breeding gilts are moved between EU Member States, they should be accompanied by a health certificate from the country of origin, signed by a veterinarian. Diseased animals cannot be transported. Operators should take preventive measures to ensure animal health during transportation. The transported animals should not pose a risk for disease introduction at the destination. Furthermore, newly introduced animals should be kept in quarantine (European Council Regulation, 2016). In Belgium, legislation on the prevention of notifiable swine diseases states that, during the first four weeks after purchasing of new animals, farmers are only allowed to transport fattening pigs to the slaughterhouse. An exception is made if the newly purchased animals are quarantined for at least four weeks and in that case, piglets can be transported to other farms and sows that need to be culled can be transported to the slaughterhouse (Royal Decree, 2014). Besides this quarantine measure, there is actually no Belgian legislation regarding the purchasing of breeding gilts nor regarding the introduction procedures of breeding gilts in pig farms.

5.2.4 MOVEMENTS OF FARM STAFF

Persons, clothing, and hands are the second most important pathogen transmission route. For example, PRRSV can be transmitted to pigs by contaminated fomites, e.g. coveralls and boots, and hands. However, the use of sanitation protocols seemed to limit the spread (Otake et al., 2002). Furthermore, a one-night downtime also prevented the spread of PRRSV by personnel and fomites (Pitkin et al., 2011).

It is important to limit the number of visitors and to apply some precautionary measures for all persons entering the pig stables to mitigate disease transmission. The European Animal Health Law emphasizes the importance of biosecurity to prevent the spread of infectious diseases to and within farms. Farm staff should acquire the appropriate knowledge and they should take action to minimize the spread of pathogens by working according to the correct working lines (European Council Regulation, 2016). Each visit to a pig farm from both farm staff and visitors should start in a hygiene lock, where farm-specific clothing and footwear can be put on and where hands can be properly washed. Furthermore, farm staff should follow a specific sequence in visiting the units with different animal categories. Younger animals are more susceptible to various pathogens due to decreased maternal immunity, while they have not yet developed a mature active immunity. This phenomenon, referred to as the immunity gap, makes piglets most susceptible around two to five weeks of age. Of course, to benefit from the passive immunity acquired by the sow, piglets must ingest sufficient colostrum during the first 24 hours of life (Fraile, 2023). On the other hand, older animals are considered to be more robust, but at the same time they may also harbor more infectious agents due to previous infections. Often these will remain unnoticed as a result of subclinical infection status. Movements or daily work should ideally be performed from young to old and from healthy to sick animals, thus according to the following sequence: (1) hygiene lock, (2) farrowing unit, (3) gestation/insemination unit, (4) nursery unit, (5) fattening unit, (6) quarantine unit, and (7) cadaver storage (Dewulf et al., 2018). Movements in the opposite direction are considered risky, as they may cause pathogen transmission. Therefore, biosecurity measures aim at separating different age groups as much as possible. If these (virtual) separations are breached in specific units or at specific time points, then the overall biosecurity goes down and the efforts made in the other units or on different time points may be nullified. Additional hygiene locks for each animal category could further reduce the risk of pathogen transmission.

Belgian legislation states that all persons, including farm staff, should change to farm-specific coverall and boots in a hygiene lock before entering the pig stables. Furthermore, hands should be washed and boots should be disinfected in a foot bath before and after visiting the stables (Royal Decree, 2020). These are external biosecurity measures, aiming to reduce or prevent the introduction of pathogens in a farm. However, there is no legislation on the working lines of persons on a pig farm, which is a part of internal biosecurity.

5.2.5 IMPACT OF BIOSECURITY ON HEALTH, PRODUCTION, ANTIMICROBIAL USE, AND ECONOMICS

Biosecurity has already been proven to positively impact a herd's health, production, AMU, and economic results. These topics will be discussed in more detail in the following paragraphs. Postma et al. (2015) investigated the perceived effectiveness, feasibility, and ROI of alternatives to antimicrobials by pig health experts from six European countries. Improved biosecurity was in the top five measures of perceived effectiveness and ROI. Mainly veterinary practitioners tended to rank biosecurity higher, while researchers and professors ranked increased diagnostics higher.

Biosecurity measures in pig herds are important to maintain animal health (Amass & Clark, 1999). Several studies have shown that biosecurity practices can be both risk or protective factors for different pathologies in swine. Ford (1995) gave recommendations regarding biosecurity to reduce the risk of spreading disease. A study on 149 French farms identified risk factors for post-weaning multisystemic wasting syndrome (PMWS), a syndrome related to porcine circovirus type 2 (PCV-2) infection. Cross-fostering of suckling piglets (internal biosecurity) increased the risk for PMWS, while rearing own breeding gilts (external biosecurity) and good cleaning procedures including a drying period of more than five days (internal biosecurity) were protective measures (Rose et al., 2003). A French study on 95 farrow-to-finish farms identified a high stocking density in the nursery unit as a risk factor for *Lawsonia intracellularis* infection (Fablet et al., 2006). Another study surveyed biosecurity and management practices on 421 pig farms. The study also described biosecurity recommendations to reduce disease (Ribbens et al., 2008). A French study on 208 farrow-to-finish and 109 fattening pig herds identified practices related to *Salmonella* prevalence in slaughter pigs. Good practices related to loading of the fattening pigs and improved cleaning and disinfection protocols reduced *Salmonella* prevalence (Corrégé et al., 2009). In a study on 108 Portuguese farrow-to-finish pig herds, a tool was developed for predicting the *Salmonella* status on farms; and herds with poor biosecurity were more likely to test positive for *Salmonella* (Baptista et al., 2010). The biosecurity status is often associated with a lower incidence of antimicrobial treatment. Frequency of treatment can be a proxy for disease incidence, thus it can be assumed that a higher biosecurity level results in healthier animals (Laanen et al., 2013; Postma et al., 2016b). A Belgian study showed that pig farmers were aware that biosecurity measures are important to reduce disease on their farms and the herd veterinarian was indicated as main source of information regarding biosecurity (Laanen et al., 2014). Although there are some studies indicating biosecurity measures as risk or protective factors for disease, there are only few intervention studies investigating the impact of improved biosecurity on health. An improved biosecurity was shown to reduce problems with the following pathogens: PRRSV (Rathkjen & Dall, 2017), *M. hyopneumoniae* (Maes et al., 2008), *Salmonella* spp. (Andres & Davies, 2015; Fraser et al., 2010; Gotter et al., 2012), and *Yersinia* spp. (Vanantwerpen et al., 2017).

For PRRSV control in a herd, The Swiss Cheese Model is suggested (Figure 12). This metaphor is used in risk management to understand how multiple layers of protection, represented as slices of cheese, can help mitigate the spread of PRRSV on a farm. No layer of defense is perfect and the potential vulnerabilities are represented by the holes in the layers. However, by having multiple layers of defense, the holes in one layer can be covered by the strengths in others. To minimize the spread of PRRSV on a farm, ten specific biosecurity measures are suggested (Boehringer Ingelheim, 2023).

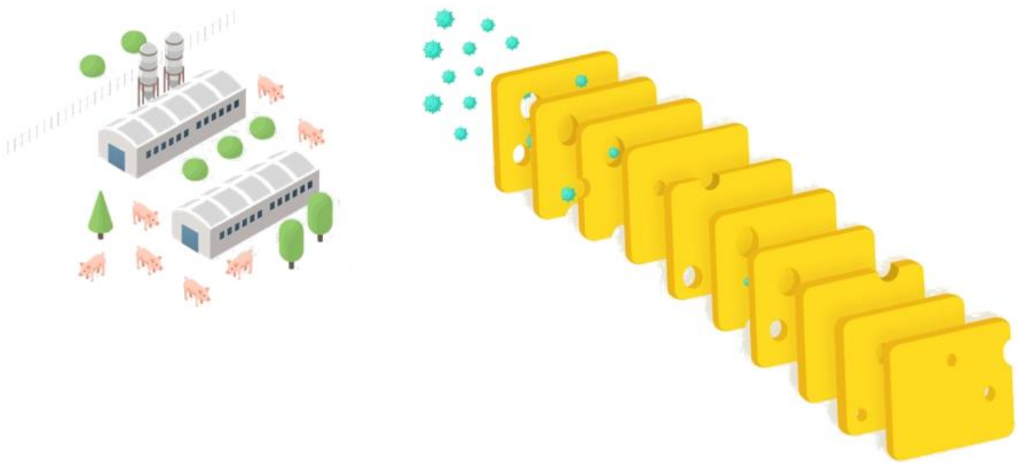

Figure 12. The Swiss Cheese Model – a multi-layered risk reduction approach (Boehringer Ingelheim, 2023).

Biosecurity has also already proven its worth in terms of production parameters in the farrowing unit, such as number of liveborn piglets and weaned piglets per sow per year; but also on production parameters of the fattening pigs, namely on average daily gain (ADG), feed conversion ratio (FCR), and mortality. The number of liveborn piglets increased by applying the all-in/all-out principle (Dors et al., 2013) or by repopulating a farm with minimum diseased breeding gilts (Nevrkla et al., 2014). The latter also lead to a decrease of the number of stillborn piglets, a decrease of the pre-weaning mortality, and consequently to an increase in the number of weaned piglets per sow per year (Nevrkla et al., 2014). Also the all-in/all-out principle and an overall good (external) biosecurity lead to an increase in the number of weaned piglets per sow per year (Dors et al., 2013; Postma et al., 2016b; Postma et al., 2017). Good biosecurity can also be beneficial for the ADG and FCR (Corrégé et al., 2011; Laanen et al., 2013; Postma et al., 2017) and can cause a decrease in mortality in the fattening unit (Corrégé et al., 2011; Postma et al., 2017). On the other hand, purchasing piglets from multiple origin farms (external biosecurity) could increase fattening pigs' mortality (Maes et al., 2004).

Several studies have already demonstrated the positive impact of biosecurity on AMU, without jeopardizing production results (Table 5). The biosecurity measures are grouped per category according to Biocheck.UGent (Biocheck.UGent, 2023).

Table 5. Overview of the biosecurity measures with a positive impact on antimicrobial use in pigs.

Category of biosecurity	Biosecurity measures	References
External biosecurity	Improved external biosecurity	Collineau et al. (2014), Postma et al. (2016a), Raasch et al. (2018), Yun et al. (2021)
Purchase of breeding gilts, piglets, and semen	Quarantine > 6 weeks	Corrégé and Hémonic (2018), Lannou et al. (2012)
	Purchasing piglets from one origin farm	Fertner et al. (2015)[a]
Transport of animals, removal of carcasses and manure	Disinfection of the loading area	Corrégé and Hémonic (2018), Lannou et al. (2012)
Supply of feed, water, and equipment	Good drinking equipment	Stygar et al. (2020)[b]
Visitors and farm staff	Presence of a hygiene lock	Dohmen et al. (2017)
	Access check of visitors and farm staff	Raasch et al. (2018)
Vermin and bird control	Professional pest control	Dohmen et al. (2017), Raasch et al. (2020)
Location of the farm	Distance to next pig farm > 500 meter	Arnold et al. (2016)
Internal biosecurity	Improved internal biosecurity	Collineau et al. (2014), Collineau et al. (2017a), Laanen et al. (2013)
Disease management	Measures related to disease management (i.e. disease control, use of hospital pens, handling of diseased animals)	Laanen et al. (2013)
Farrowing and suckling period	Measures related to farrowing and suckling period (i.e. washing of sows, cross-fostering, handling of piglets)	Laanen et al. (2013), Raasch et al. (2020)
Nursery unit	All-in/all-out in the nursery unit	Corrégé and Hémonic (2018), Dupont et al. (2017), Fertner et al. (2015)[a], Lannou et al. (2012)
	Solid partitions in nursery pens to limit contact	Corrégé and Hémonic (2018), Lannou et al. (2012)
Fattening unit	All-in/all-out in the fattening unit	Corrégé and Hémonic (2018), Dupont et al. (2017), Lannou et al. (2012)
	Stocking density	Stygar et al. (2020)[b]
Measures between compartments, working lines, and use of equipment	Working lines	Corrégé and Hémonic (2018), Lannou et al. (2012), Raasch et al. (2020)
Cleaning and disinfection	Good cleaning and disinfection	Dupont et al. (2017), Fertner et al. (2015)[a], Raasch et al. (2018), Raasch et al. (2020), Stygar et al. (2020)[b]
Overall biosecurity	Improved biosecurity	Postma et al. (2017)

All studies were performed on farrow-to-finish farms, except for:

[a] this study was performed on a nursery pig farm;

[b] this study was performed on a fattening pig farm.

Improved biosecurity has also been related to improved farm profitability. Two French studies examined the relationship between biosecurity and the economical performances of farms. The first study was performed on 166 farrow-to-finish pig farms. The impact of good biosecurity and management practices was estimated to result in a benefit of € 182 per sow per year compared to farms with unfavorable practices (Corrégé et al., 2011). The second study focused on biosecurity practices only and was performed on 77 farrow-to-finish farms. Farms with a higher overall biosecurity showed a benefit of € 202 per sow per year compared to farms with a low overall biosecurity (Corrégé et al., 2012). An economic evaluation based on the results of the study of Postma et al. (2017) showed that a benefit of € 2.67 per fattening pig per year was gained after biosecurity interventions (Rojo-Gimeno and Postma et al., 2016). A similar study was conducted in four European countries, where a reduction in AMU was achieved by herd-specific interventions, including biosecurity, without a negative impact on production parameters. The median change in net farm profits was estimated at € 4.46 and € 1.23 per sow per year in Belgian and French farms, respectively (Collineau et al., 2017b).

5.3 VACCINATION

Vaccines may have great value in preventing diseases and there are many different commercial vaccines against both bacterial and viral pathogens. Vaccination has been identified as one of the most feasible alternatives for antimicrobial treatment (Postma et al., 2015). Even though there is an initial cost of purchasing vaccines, vaccination may have a beneficial ROI (Maes et al., 2003; Wittum & Dewey, 1996; Young et al., 2011). Several studies have confirmed that AMU could be reduced by increased vaccination. Sometimes vaccination against one pathogen was investigated, such as *M. hyopneumoniae* (Mateusen et al., 2001), *A. pleuropneumoniae* (Del Pozo Sacristán et al., 2014), *L. intracellularis* (Adam, 2009; Bak, 2011; Bak & Rathkjen, 2009; Coube et al., 2012), or PCV-2 (Aerts & Wertenbroek, 2011; Brockhoff et al., 2009; Coube et al., 2012; Raith et al., 2016). In other studies, vaccination against multiple pathogens was suggested as part of a herd-specific action plan to prevent disease and to reduce AMU (Dupont et al., 2017; Postma et al., 2017).

5.4 FEED (ADDITIVES)

Optimal feed is necessary for growth in pigs, but also to avoid problems with the intestinal health, e.g. weaning diarrhea and swine dysentery, and lameness, which can lead to an increased AMU. Furthermore, the feed is important for the pig's immunity. There are several elements to consider, namely energy and protein levels, vitamins and minerals, fiber, form and structure, but also feeding strategies (Millet & Everaert, 2023). Improved feed quality was perceived to have the highest impact on reducing AMU (Speksnijder et al., 2015b).

Prebiotics, probiotics, synbiotics, or organic acids can be supplemented to the feed. Prebiotics are fermentable components of feed, such as oligosaccharides or dietary fibers, that modify the intestinal microbiota to improve the animal's health (Gibson et al., 2004). Probiotics are live microbials that are supplemented to the feed, such as *Lactobacillus*, *Bacillus*, *Bifidobacterium*, or yeasts (Stein & Kil, 2006). Probiotics can change the balance of the microbiota in the intestinal tract and compete with pathogenic bacteria, resulting in improved intestinal health. Synbiotics, the combination of pre- and probiotics, have an even greater beneficial effect on the microbiota (Malik et al., 2019). Also organic acids can be supplemented to the feed. Organic acids can decrease the pH, which makes it difficult for pathogens to survive (Allen et al., 2013). Also phytogenic feed additives, i.e. herbs or plants, are sometimes used in the pig's feed. Several studies have shown that these additives can also have an antimicrobial action (Michiels et al., 2009; Mohammadi Gheisar & Kim, 2018).

Since late 2013 and especially in 2014, zinc oxide has been used medicinally to prevent post-weaning *E. coli* diarrhea in nursery pigs. Zinc oxide was in the top five of perceived feasibility and ROI as alternative for antimicrobials, especially by veterinary practitioners and nutritionists (Postma et al., 2015). However, zinc oxide burdens the environment because zinc as heavy metal is excreted in the environment (Agence Nationale de Sécurité Sanitaire Alimentation Environnement Travail (ANSES), 2013) and it is also related to potential selection of AMR (Medardus et al., 2014). Therefore, it was later decided at the European level that zinc oxide could no longer be used medicinally by the summer of 2022 (European Commission, 2017a). In Belgium, the registration of the only veterinary medicinal product with zinc oxide (Gutal®) expired in 2019. It was decided that the stock of this product could still be used, but the use should be phased out by the end of 2020, and it could no longer be used from 2021 onwards. There were fears that this would cause an increase in AMU (AMCRA, 2018), but that has not been the case. Zinc may still be used as a feed additive, as in this case, the concentrations are much lower.

5.5 STABLE CLIMATE AND HOUSING

There are several elements of stable climate that significantly impact animal health, namely ventilation, temperature, and air quality. Suboptimal air quality and low temperatures can lead to different health and welfare problems such as respiratory disease or diarrhea in pigs, both leading to increased AMU (McEwen & Fedorka-Cray, 2002). High ammonia concentrations can irritate the respiratory mucosa and can facilitate respiratory infections, leading to an increased AMU (Albernaz-Gonçalves et al., 2022). Also particulate matter (PM) affects the respiratory health of pigs and increasing PM concentrations lead to increasing odds of pneumonia and pleurisy lesions (Michiels et al., 2015). A study in Belgium and The Netherlands showed that veterinary practitioners believed suboptimal climate was the main cause of high AMU (Postma et al., 2016c).

For the housing, pen design, stocking density, group size, flooring, and access to feed and water can affect animal health. Insufficient environmental enrichment can lead to frustration in pigs, leading to inappropriate behavior such as tail or ear biting, resulting in infections and abscesses. Also aggression, e.g. by mixing pigs, can lead to skin lesions. Both conditions can subsequently lead to an increased AMU. A high stocking density causes stress in pigs, resulting in an increased sensitivity for infection and also an increased excretion of pathogens. Furthermore, a high stocking density causes a significant increase of pathogen transmission (Dewulf et al., 2018) and it could also result in joint infections or tail biting, which can cause an increased AMU (Albernaz-Gonçalves et al., 2022). These examples demonstrate the importance of optimizing stable climate and housing to prevent health problems and the resulting increased AMU.

5.6 COACHING

In order to reduce AMU on pig farms, a farm-specific approach is important and the herd veterinarian plays a key role (Alarcon et al., 2014). Traditionally, the herd veterinarian acts as an advisor. When problems occur, farmers contact the veterinarian, who subsequently provides advice. The same approach could be used for reducing AMU; however, previous studies have shown the importance of the farmer's mindset and social psychology in this context (Visschers et al., 2015). To guide farmers towards more prudent AMU practices, long-term changes in their habits are essential, and coaching could be the way to accomplish this. Coaching differs from traditional advising, because the coach helps finding the farmer's motivation to obtain a behavioral change.

The Theory of Planned Behavior is a widely used psychological model that can explain human behavior in various domains, including livestock farming (Figure 13). This theory suggests that human behavior is influenced by three key factors: attitudes, subjective norms, and perceived behavioral control. Attitudes refer to a person's overall evaluation of a behavior, which can be positive or negative. The subjective norms refer to the perceived social pressure or influence from others to engage in a particular behavior. If a farmer perceives that their social network expects them to adopt certain practices, it can influence their behavior. Perceived behavioral control refers to a person's belief in their ability to perform a behavior (Ajzen, 1991).

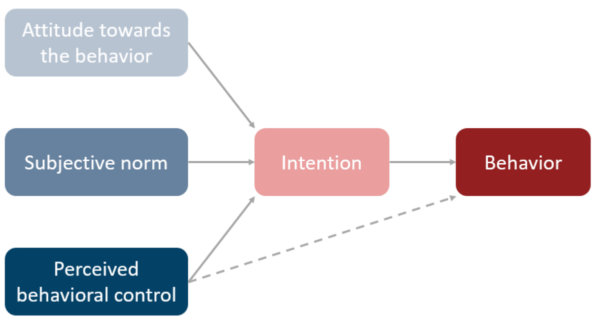

Figure 13. Visual representation of the Theory of Planned Behavior (after Ajzen, 1991).

Different studies already applied coaching in pig farms. Sixty-one Belgian farrow-to-finish pig herds were coached to reduce their AMU by herd-specific intervention plans (Postma et al., 2017). A similar study was performed on 68 farrow-to-finish pig herds in Belgium, France, Germany, and Sweden (Raasch et al., 2020). Both studies emphasized the importance of coaching and the cooperation between the farmer, the herd veterinarian, and an external coach for a good compliance of implemented measures. A decent follow-up of the situation and specific suggestions for improvement, combined with information on the risks when certain practices would not be performed correctly, accompanied by (positive) feedback seemed crucial to maintain high levels of motivation.

To quantify the effect of coaching, the ADKAR® model is suggested. ADKAR® is an acronym for five elements that are important for change to succeed and the model was adapted for use in veterinary medicine. The model can specifically be used for livestock antimicrobial stewardship (Table 6) (Caekebeke, 2021; Houben et al., 2020). For each element, a score from 1 to 5 is given, with 1 representing the lowest and 5 the highest score. If an element scores 3 or less, change can be blocked (Hiatt, 2006).

Table 6. The ADKAR® model for livestock antimicrobial stewardship (adapted from Houben et al., 2020).

ADKAR	Description
Awareness	Represents the awareness that AMU in livestock production should be reduced, since this is a risk for introduction of antimicrobial resistant bacteria to humans and animals.
Desire	Represents the personification of the awareness. “Does the farmer himself want to reduce AMU in his farm?”
Knowledge	Represents the knowledge and skills of the farmer to implement measures to improve health and to reduce the need for antimicrobial treatment.
Ability	Represents the implementation phase of the change. Will or is the farmer implement(ing) changes in management or working methods? (Topics for change are: feed, management, climate, working methods, etc.).
Reinforcement	Represents the sustainability of change. An active positive reinforcement is necessary to sustain change.

When Houben et al. (2020) adapted the ADKAR® model for use in veterinary medicine, they immediately profiled 26 poultry and 28 pig farmers from Belgium and The Netherlands. It was expected that in different farms successful change would be limited by lack of different elements (score ≤ 3) from the ADKAR® model: awareness (40 %), desire (54 %), knowledge (70 %), ability (52 %); and 83 % of the farmers scored 3 or less for at least one of the elements. In these farms, the ADKAR®-elements should first be properly addressed, before coaching could focus on technical veterinary advice on farm management. However, this study did not assess an immediate association between ADKAR® profiling scores and AMU at farm or country level .

Caekebeke et al. (2021) combined ADKAR® profiling and coaching on 30 broiler farms in Belgium and The Netherlands. Different improvements were made on the farms, namely an increased ADKAR® score, improved biosecurity levels, and a reduction in AMU without compromising performance. However, no significant association was found between higher ADKAR® scores and lower AMU and further research is needed to assess the role of ADKAR® in reducing AMU in livestock production.

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CHAPTER 2 | SCIENTIFIC AIMS

Worldwide, pigs are one of the most commonly raised animals for meat production. To meet the rising demand for animal protein, livestock production has been intensified during the past decades. This intensification was accompanied by increased stocking density, leading to a more efficient pathogen transmission between animals, resulting in increased antimicrobial use (AMU). Unfortunately, AMU results in selection pressure for antimicrobial resistance (AMR). In Europe and Belgium, pig production is subject to strict regulations regarding the use of antimicrobials and other veterinary drugs. Also quality labels and concepts like Raised Without Antibiotics (RWA) have found their way into pig production. Nonetheless, an RWA program is not yet established in Belgium. Furthermore, the specific characteristics of farms that can raise pigs without antimicrobials are unknown.

Collecting objective data is crucial for the development of reduction strategies. Of 31 European countries participating in the ESVAC project, Belgium is the 10th highest user of veterinary antimicrobials. Although AMU in pigs is decreasing over the years, there is still much room for improvement. In order to reduce AMU, animal health should be improved. The focus should be on disease prevention, e.g. by increasing biosecurity levels in farms. To do so, the situation should be assessed first. Afterwards, awareness can be created and the situation can be improved by increasing knowledge on the subject. Live animals include the highest risk of disease transmission, yet it is not known if Belgian farms comply to the optimal introduction procedures of breeding gilts upon purchase as described in literature. Another important disease transmission route is people. Farm staff should stick to specific working lines, i.e. from young to old, when performing the work on the farm. However, to date there is no knowledge on the movements of farm staff on pig farms.

The general aim of this thesis was to create awareness on biosecurity, to improve animal health, to reduce antimicrobial use, and to investigate the feasibility of RWA on pig farms.

The specific objectives were:

1. to investigate the **procedures upon purchase of breeding gilts** and the compliance to the optimal introduction procedures;

2. to assess **movements of farm staff** on pig farms, to assess risky movements, and to investigate whether movements differed according to time (week of the batch farrowing system and weekday vs. weekend) and unit (farrowing, gestation/insemination, nursery, and fattening unit);

3. to describe the criteria for a Belgian **Raised Without Antibiotics** program, to evaluate whether farms could achieve and maintain this status, and to identify possible differences between RWA and non-RWA farms.

CHAPTER 3 | PURCHASING POLICY, QUARANTINE AND ACCLIMATION PRACTICES OF BREEDING GILTS ON BELGIAN PIG FARMS

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ABSTRACT

The breeding population is very important in pig herds for productivity, health, and profitability. Replacement of breeding animals can be accomplished by own rearing of breeding gilts or by purchasing them. Purchasing breeding gilts is a hazardous event in terms of biosecurity and introduction of pathogens into a farm. However, in literature little is known about gilt introduction in a herd. The present study investigated the introduction procedures of purchased breeding gilts in Belgian pig herds and the compliance of these herds to the optimal introduction procedures. A questionnaire consisting of twenty questions related to farm characteristics (*n* = 2), purchasing policy (*n* = 6), quarantine period (*n* = 5), and acclimation practices (*n* = 7) was designed and 68 farms completed the questionnaire during an on-farm interview. The median (min. – max.) number of sows on the farms was 300 (85 – 2500). Fifty-seven percent of the farms purchased breeding gilts and there was a lot of variation in the frequency of purchase and the age at which gilts were purchased. On 95 % of those farms, a quarantine unit was used and on most of these farms the quarantine was located on the farm itself (internal quarantine). The median (min. – max.) duration of the quarantine period was 42 (14 – 140) days. The most common acclimation practice was vaccination against porcine parvovirus (96 %) and *Erysipelothrix rhusiopathiae* (94 %), although in some farms exposure of gilts to farm-specific micro-organisms was done by providing feces from suckling piglets (18 %) and bringing gilts in contact with sows that will be culled (16 %). Only 10 % of the farms complied with the optimal introduction procedures, i.e. purchasing policy, quarantine building and quarantine management. This study showed that in many farms, practices related to purchasing, quarantine and acclimation could be improved to maintain optimal biosecurity.

INTRODUCTION

The breeding population is very important in pig herds for productivity, health, and profitability (Patterson & Foxcroft, 2019). Replacement of breeding animals can be accomplished by own rearing of breeding gilts or by purchasing them. Purchasing of breeding gilts might lead to faster improvement of genetic potential, but it includes the risk of pathogen introduction in a farm (Pritchard et al., 2005). For 14 bacteria and 10 viruses causing diseases in swine, transmission by direct contact has been described, i.e. transmission by secretions and excretions of live animals or cadavers (Filippitzi et al., 2018). For example, the transmission rate of *Actinobacillus pleuropneumoniae* is estimated to be 10 times higher for direct contact in comparison to indirect transmission (Tobias et al., 2014). Purchasing breeding gilts was found to be a risk factor for seroprevalence of *Mycoplasma hyopneumoniae* in slaughter pigs on farrow-to-finish pig herds (Maes et al., 1997). Therefore, purchasing breeding gilts is a hazardous event in terms of the introduction of new pathogens into a farm. Several factors should be taken into account, such as frequency of purchase, number of animals purchased, number of origin herds, the transport vehicle, and the health status of the origin farms (Neumann & Hall, 2019).

Placing purchased animals in a quarantine unit aims (1) to reduce the risk of pathogen introduction into the farm, and (2) to facilitate the introduction of the animals into the herd by means of acclimation. During the quarantine period, pigs can be observed for the presence of clinical signs and be tested for the presence of pathogens. Acclimation practices such as vaccination against several pathogens and exposure to live animals (e.g. pigs or sows before culling), can protect newly purchased animals against pathogens circulating on the farm (Garza-Moreno et al., 2017; Pritchard et al., 2005).

Based upon guidelines described in literature, optimal introduction procedures in terms of good biosecurity can be divided into three main categories, namely purchasing policy, quarantine building, and quarantine management (Dewulf et al., 2018). From a biosecurity viewpoint, purchasing animals constitutes a risk that can only be minimized, but not completely eliminated. If animals need to be purchased, the farmer could pay attention to the following items to minimize the risk of pathogen introduction: the pigs should always originate from the same farm, the health status of the origin farm should be higher than or equal to the farm, and strict hygiene measures should be implemented for the transport vehicle. Once the newly purchased pigs arrive on the farm, there are some requirements for the building and the management of the quarantine unit. A quarantine unit should be present and ideally it should be completely isolated from the other animals, with a separate entrance and hygiene lock. The all-in/all-out principle should be practiced, so that new gilts can only arrive when the quarantine unit is completely empty. The duration of the quarantine period should be a minimum of 28 days (Dewulf et al., 2018).

The present study investigated the introduction procedures of gilts on Belgian pig farms as a first step to optimize the health management of breeding gilts. We focused on purchasing policy, quarantine period, and acclimation practices. The results were compared to the optimal situation, to determine to what extent these practices are in line with the recommendations for optimal introduction procedures (Dewulf et al., 2018).

MATERIALS AND METHODS

DESIGN OF THE QUESTIONNAIRE

The questionnaire was designed based on the principles outlined in the chapter ‘Questionnaire Design’ from the book ‘Veterinary Epidemiologic Research’ (Dohoo et al., 2014). The questionnaire was kept short and not too complex, to lower the response burden and thus increase the response rate. To identify confusing or ambiguous questions, the questionnaire was pre-tested and evaluated by veterinarians of the Unit of Porcine Health Management (Faculty of Veterinary Medicine, Ghent University) (n = 4), veterinarians from Animal Health Care Flanders (n = 3), and a pig practitioner (n = 1). Based on their feedback, some final changes were made.

The questionnaire was limited to 20 questions and some questions from Biocheck.UGent, a risk-based biosecurity scoring tool, were included (Biocheck.UGent, 2023). First, some general information was asked, such as herd size and the batch farrowing system. Further, the questionnaire was divided into three parts namely purchasing policy, quarantine period, and acclimation practices of breeding gilts, each consisting of six, five and seven questions, respectively, regarding the past year. Most questions were multiple choice questions, but sometimes 'fill-in-the-blank' questions were used to request additional information or to collect numerical data. A checklist was used for some questions. In the case where breeding gilts were not purchased, subsequent questions related to purchasing policy and quarantine were skipped. The different questions of the questionnaire are shown in Tables 1, 2, 3, and 4.

For the entire questionnaire, except for the general information, there were five questions where numerical data were collected (Table 4). There were three variables for the part on purchasing policy: years of cooperation with the same origin farm, frequency of purchase, and age of the purchased gilts; one variable for the part on the quarantine period: duration of quarantine; and one variable on the acclimation practices: stocking density of the breeding gilts in group housing.

The questions on the purchasing policy of breeding gilts are presented in Table 1. Table 2 shows the questions on the quarantine period of breeding gilts. If farmers indicated the presence of a quarantine unit, they had to clarify whether the quarantine unit was located externally, i.e. on a different site away from the farm, or internally, i.e. on the same site as the farm. They had to specify the type of stable as well; an isolated stable meant that the air volume and manure pit were completely separate. The all-in/all-out principle meant that new gilts could only enter the stable after the previous batch had moved to a new compartment. A separate hygiene lock was defined as a room where people could change their coverall and boots and wash their hands before entering the quarantine.

There were different questions for collecting information on acclimation practices (Table 3). The first one was related to the vaccination strategies that were used, e.g. age at vaccination, where vaccinations were given (origin farm, purchasing farm), and the product used. Farmers who raised their own breeding gilts could also indicate which vaccinations were applied in the rearing unit. Pathogens against which vaccination is common and/or for which commercial vaccines are available were considered. The information on vaccination, including vaccination strategies, were classified in seven categories: no vaccination, one vaccination at the origin farm, one vaccination in the quarantine unit, more than one vaccination at the origin farm, more than one vaccination in the quarantine unit, combination of vaccinations at the origin farm and the quarantine unit, and vaccination without further details. Other acclimation practices, monitoring for pathogens, and questions on housing conditions of the breeding gilts, were included as well (Table 3).

DISTRIBUTION OF THE QUESTIONNAIRE

Belgian pig farmers were contacted and visited by veterinarians of the Unit of Porcine Health Management (*n* = 6), a veterinarian at Animal Health Care Flanders (*n* = 1), or by pig practitioners (*n* = 4). The pig farmers were able to participate voluntarily and the questionnaires were filled out during an on-farm interview linked to routine farm visits. Therefore, the selected farms are a convenience sample. The answers given by the farmer were considered to be accurate and were not verified. For most herd visits done by the Unit of Porcine Health Management and Animal Health Care Flanders, the herd veterinarian was present, which enhances the validity of the answers. Questionnaires were collected from 1 October 2019 until 31 March 2020.

ANALYSIS OF THE DATA

Descriptive statistics were performed for both the continuous and the categorical variables of the questionnaire. For the continuous variables, the median, minimum, and maximum values were determined. For the categorical variables, percentages were calculated. No categories were deleted; however, some categories were merged to simplify complex outcomes. Normality distribution was analyzed graphically via histograms and Q-Q plots.

A non-parametric Mann-Whitney U test was used to analyze potential differences between farms for the not normally distributed data, i.e. herd size and duration of the quarantine period. The Levene's test was used for analyzing equality of variances. A parametric independent samples t-test was used to analyze potential differences between farms for the normally distributed data, i.e. frequency of purchasing breeding gilts and number of different vaccinations in gilts. A Chi-Square test of independence was used to assess differences between categorical variables. *P*-values below 0.05 were considered statistically significant. Statistical analyses were mainly performed using IBM® SPSS® Statistics for Windows Version 24 (IBM Corp., Armonk, N.Y., USA).

The agreement between the observed measures and the described optimal procedures was analyzed by means of a categorical variable decision tree. First, it was checked whether they applied all three purchasing principles, i.e. the same origin farm, high health status of origin farm, and requirements for the transport truck. Second, the quarantine building was checked, i.e. having a separate air volume and having a separate hygiene lock for the quarantine unit. Third, the quarantine management was evaluated, i.e. using the all-in/all-out principle and having a quarantine duration of minimum 28 days. If all previously mentioned procedures were applied, the farm was considered to comply with the optimal introduction procedures as described in the Introduction.

RESULTS

PARTICIPATING PIG HERDS

All contacted pig farmers (*n* = 68) completed the questionnaire. All farms were located in Flanders. The median (min. – max.) number of sows on the farms was 300 (85 – 2500). The 1-, 2-, 3-, 4- and 5-week batch farrowing system were used on 14 % (10/68), 6 % (4/68), 31 % (21/68), 37 % (25/68), and 12 % (8/68) of the farms, respectively.

PURCHASING POLICY

The results on the purchasing policy of breeding gilts are shown in Tables 1 and 4. Breeding gilts were purchased on 57 % (39/68) of the farms, while the remaining 43 % (29/68) reared their own breeding gilts. Ninety-seven percent (38/39) of the purchasing farms worked with the same origin farm each time they purchased breeding gilts. Only 47 % (18/38) of the latter indicated the duration of their cooperation with the same origin farm, which was in median (min. – max.) 5 (1 – 12) years.

Table 1. Results of the categorical variables related to purchasing policy of breeding gilts in pig farms.

Variable	n	%
Are breeding gilts purchased? (*n* = 68)		
Yes	*39*	*57*
No	*29*	*43*
Do you always work with the same origin farm or do you cooperate with multiple? (*n* = 39)		
Always same origin farm	*38*	*97*
Multiple origin farms	*1*	*3*
Do you have information on the health status of the origin farm? (*n* = 39)		
Higher than or equal to own farm	*31*	*79*
Lower than own farm or not known	*8*	*21*
Are there hygienic requirements for the transport truck of purchased breeding gilts? (*n* = 39)		
Yes	*25*	*64*
No	*14*	*36*

Seventy-nine percent (31/39) of the purchasing farmers claimed a health status of the origin farm that was higher than or equal to their own farm, whereas on 21 % (8/39) of the purchasing farms, the health status was lower or the farmers did not know the health status of the origin farm. There were specific requirements for transport on 64 % (25/39) of the purchasing farms. The most common requirement was that the truck was exclusively used for transport of breeding gilts (44 %, 11/25), followed by a cleaned and disinfected truck (28 %, 7/25), and transport each time from one origin farm to only one purchasing farm (24 %, 6/25). Other requirements for gilt transport were that it had to be done as first job of the day

(16 %, 4/25) or specifically as first job on Monday (12 %, 3/25). The median (min. – max.) frequency of purchasing breeding gilts was 6 (1 – 13) times per year. The median (min. – max.) age of the purchased gilts was 24 (9 – 37) weeks.

QUARANTINE PERIOD

The results related to the quarantine period are shown in Tables 2 and 4. On 95 % (37/39) of the purchasing farms, a quarantine unit was present. The quarantine unit had an external location in 3 % (1/37) of the cases and an internal location in 97 % (36/37) of the cases. When an external quarantine was used, the gilts were subsequently housed in an internal quarantine unit before they joined the sow population in the herd. The gilts were isolated in a separate stable on 62 % (23/37) of the farms, whereas on 35 % (13/37) of the farms, the gilts were housed in a separate compartment within a stable. Two of the latter farmers specified that there was a separate manure pit and one farmer reported that there was a separate ventilation system in the quarantine compartment. Hence, 68 % (25/37) of the farms had a quarantine unit with a separated air volume, i.e. the farms with an external quarantine unit (*n* = 1), the farms with an isolated stable (*n* = 23), and the farms that indicated separate ventilation in the quarantine compartment (*n* = 1). For the farms with a quarantine unit (*n* = 37), the all-in/all-out principle was used for the quarantine unit on 86 % (32/37) of the farms and a separate hygiene lock for the quarantine unit was present on 54 % (20/37) of the farms. The median (min. – max.) duration of the quarantine period was 42 (14 – 140) days.

Table 2. Results of the categorical variables related to the quarantine period of breeding gilts in purchasing farms.

Variable	n	%
Is a quarantine unit present at the farm? (*n* = 39)		
Yes	*37*	*95*
No	*2*	*5*
What is the location of the quarantine unit? (*n* = 37)		
External – followed by internal quarantine	*1*	*3*
External – adding gilts immediately to the herd	*0*	*0*
Internal – isolated stable	*23*	*62*
Internal – separate compartment within a stable	*13*	*35*
Internal – together with other pigs on the farm	*0*	*0*
Is the all-in/all-out principle used in the quarantine unit? (*n* = 37)		
Yes	*32*	*86*
No	*5*	*14*
Do you have a separate hygiene lock for the quarantine unit? (*n* = 37)		
Yes	*20*	*54*
No	*17*	*46*

ACCLIMATION PRACTICES

Table 3 shows the frequency of vaccination of breeding gilts against different pathogens. The vaccination strategies for each pathogen are shown in Figure 1. Furthermore, two farmers vaccinated the gilts with an autogenous vaccine against *Streptococcus suis* and on one of those farms, *Staphylococcus hyicus* was included in the vaccine as well. The median (min. – max.) number of pathogens against which gilts were vaccinated was 7 (2 – 12).

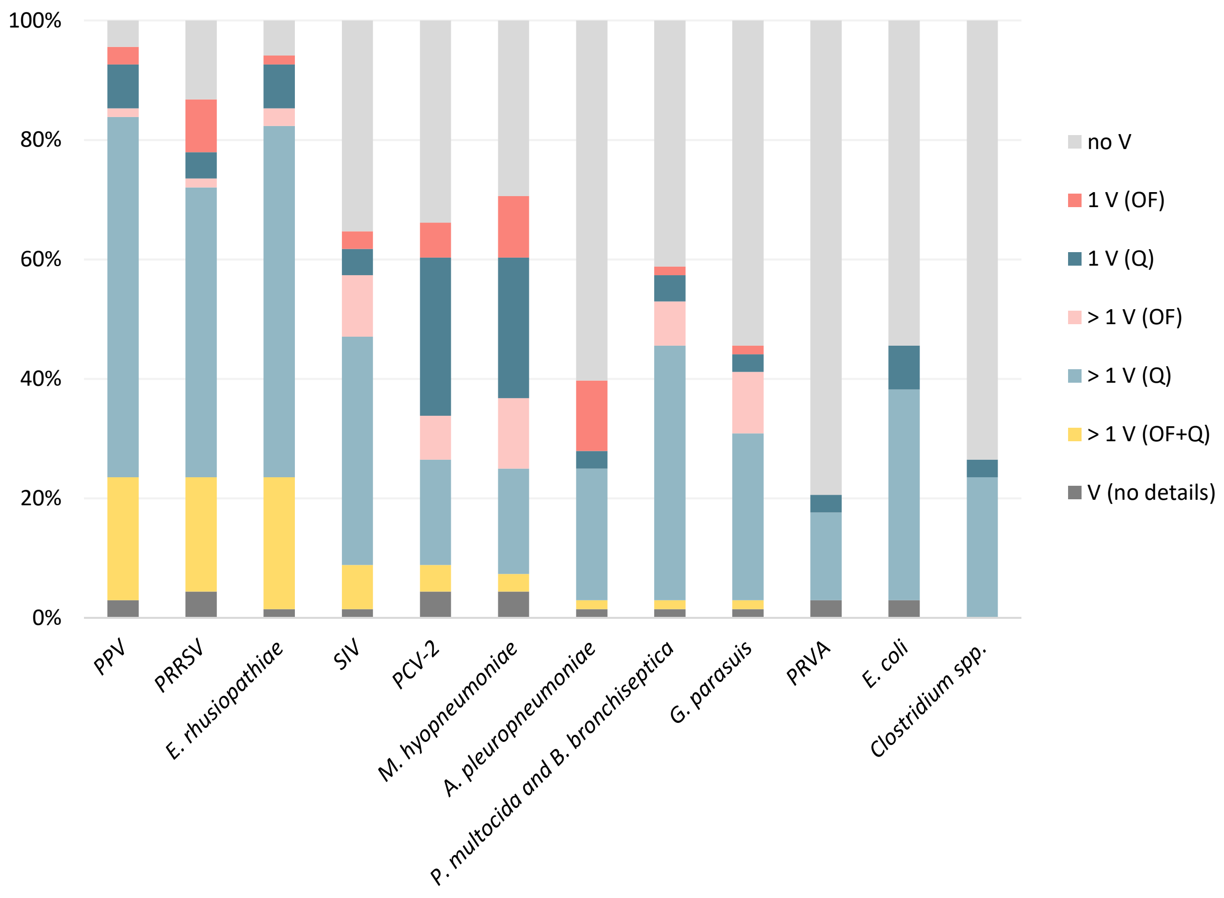

Figure 1. Vaccination strategies [a] (*n* = 7) used on all the farms (*n* = 68) for the different pathogens [b] (*n* = 12).

[a] no V: no vaccination; 1 V (OF): one vaccination at the origin farm; 1 V (Q): one vaccination in the quarantine unit; > 1 V (OF): more than one vaccination at the origin farm; > 1 V (Q): more than one vaccination in the quarantine unit; > 1 V (OF+Q): combination of vaccinations at the origin farm and the quarantine unit; V (no details): vaccination without further details

[b] The pathogens were grouped based on whether they affected mainly reproductive performance (porcine parvovirus (PPV), porcine reproductive and respiratory syndrome virus (PRRSV) and *Erysipelothrix rhusiopathiae*), or respiratory (swine influenza virus (SIV), porcine circovirus type 2 (PCV2), *Mycoplasma hyopneumoniae*, *Actinobacillus pleuropneumoniae*, *Pasteurella multocida* and *Bordetella bronchiseptica*, and *Glaeserella parasuis*) or intestinal health (porcine rotavirus type A (PRVA), *Escherichia coli* and *Clostridium* spp.).

Table 3. Results of the categorical variables related to the acclimation practices of breeding gilts in pig farms.

Variable	**n**	**%**
Against which pathogens are the breeding gilts vaccinated? [a,b] (*n* = 68)		
PPV	*65*	*96*
PRRSV	*59*	*87*
E. rhusiopathiae	*64*	*94*
SIV	*44*	*65*
PCV-2	*45*	*66*
M. hyopneumoniae	*48*	*71*
A. pleuropneumoniae	*27*	*40*
P. multocida and B. bronchiseptica	*40*	*59*
G. parasuis	*31*	*46*
PRVA	*14*	*21*
E. coli	*31*	*46*
Clostridium spp.	*18*	*26*
Which acclimation practices are being used? Contact with… [a] (*n* = 68)		
Sows that will be culled	*11*	*16*
Placenta tissue	*6*	*9*
Feces from suckling piglets	*12*	*18*
Feces from nursery pigs	*2*	*3*
Feces from piglets with diarrhea	*1*	*1*
Other	*21*	*31*
None	*29*	*43*
Are breeding gilts monitored for specific pathogens? [a] (*n* = 68)		
Yes, for Brachyspira hyodysenteriae only	*2*	*3*
Yes, for other pathogens than B. hyodysenteriae	*7*	*10*
Yes, for B. hyodysenteriae and other pathogens	*2*	*3*
No	*57*	*84*
How are breeding gilts housed? (*n* = 68)		
Individual housing	*5*	*7*
Group housing	*56*	*82*
Combination of individual and group housing	*7*	*10*

[a] Farmers could give several answers to these questions, therefore the sum of the percentages can exceed 100 %.

[b] The vaccinations were grouped based on whether the pathogen affected mainly reproductive performance (porcine parvovirus (PPV), porcine reproductive and respiratory syndrome virus (PRRSV) and *Erysipelothrix rhusiopathiae*), or respiratory (swine influenza virus (SIV), porcine circovirus type 2 (PCV-2), *Mycoplasma hyopneumoniae*, *Actinobacillus pleuropneumoniae*, *Pasteurella multocida* and *Bordetella bronchiseptica*, and *Glaeserella parasuis*) or intestinal health (porcine rotavirus type A (PRVA), *Escherichia coli* and *Clostridium* spp.).

Fifty-seven percent (39/68) of the farms used one or more acclimation practices (Table 3). Giving feces from suckling piglets to the gilts was used in 18 % (12/68) of the farms, followed by housing sows that will be culled in the same compartment as the breeding gilts (16 %, 11/68). Other acclimation practices included the provision of placenta tissue (9 %, 6/68), feces from nursery pigs (3 %, 2/68), and feces from piglets with diarrhea (1 %, 1/68) to the breeding gilts. Several other acclimation practices were also used (31 %, 21/68), such as giving feces from sows, providing a burlap bag which hung first in the farrowing or nursery unit for contact with feces or oral fluids, and giving leftovers from the feeding corridor to the breeding gilts. Eighteen percent (12/68) of the farms used different combinations of acclimation methods. Seven percent (5/68) of the farms indicated that they did own rearing of gilts and that gilts were housed in a pen in the fattening or gestation unit.

Eighty-four percent (57/68) of the farms did not monitor for any pathogen during the quarantine or acclimation period (Table 3). Two farms took fecal samples to monitor *Brachyspira hyodysenteriae* via PCR testing (3 %), while other farms monitored the presence of antibodies in serum against other pathogens using ELISA (10 %, 7/68), and two farms practiced the combination of both (3 %).

Table 3 shows the information on housing of breeding gilts. Breeding gilts were housed in different ways. On 7 % (5/68) of the farms, they were kept individually, while on 82 % (56/68) of the farms, the gilts were housed in groups. Ten percent (7/68) of the farms used a combination of individual and group housing. The median (min. – max.) stocking density on the farms, where gilts were housed in groups, was 1.00 (0.75 – 5.00) m² per gilt. Farms were categorized in three different categories according to stocking density of breeding gilts: < 1 m² (8 %, 4/53), 1 – 1.5 m² (74 %, 39/53), and > 1.6 m² (19 %, 10/53).

Table 4. Results of the continuous variables in the questionnaire.

Variable	median	min.	max.
How long are you already working with the same origin farm (years)? (*n* = 18)	5	1	12
What is the frequency of purchasing breeding gilts (times per year)? (*n* = 39)	6	1	13
What is the age of the purchased breeding gilts (weeks)? (*n* = 39)	24	9	37
What is the minimum duration of the quarantine period (days)? (*n* = 37)	42	14	140
What is the stocking density of the breeding gilts kept in group housing (m²)? (*n* = 53)	1.00	0.75	5.00

DIFFERENCES BETWEEN THE HERDS

There was a statistically significant difference in herd size between farms that required hygienic measurements (median: 360 sows) and farms that did not require hygienic measurements of the transport vehicle (median: 220 sows) ($p = 0.006$). There was a statistically significant difference in duration of the quarantine period between farms that had a separate hygiene lock (median: 46 days) and farms that did not have a separate hygiene lock for the quarantine unit (median: 31 days) (p = 0.007). There was a significant increase in the frequency of purchasing breeding gilts by on average 4 times per year (95% CI: 1 to 7, p = 0.023) for farms that did not use the all-in/all-out principle (10 ± 2 times per year) compared to farms that did use the all-in/all-out principle in the quarantine unit (6 ± 3 times per year). There was a significant decrease in number of pathogens against which gilts were vaccinated by on average two pathogens (95% CI: -3 to -1, p = 0.004) for farms that reared their own gilts (6 ± 3 pathogens) compared to farms that purchased breeding gilts (8 ± 3 pathogens). No statistically significant associations were found between the purchasing of breeding gilts and the use of a batch farrowing system (χ^2 = 0.000, p = 1.000), nor between the purchasing of breeding gilts and the use of acclimation practices (χ^2 = 0.655, p = 0.418).

OPTIMAL INTRODUCTION PROCEDURES

For the purchasing farms, the compliance to the optimal introduction procedures was verified (Table 5). Fifty-four percent (21/39) of the farms applied all three principles of the purchasing policy. Ninety-five percent (37/39) of the farms housed their gilts in a quarantine unit. Thirty-eight percent (14/37) of the farms with a quarantine unit had a proper quarantine building, i.e. a stable with a separate air volume (external or internal location) with a separate hygiene lock. Eighty-one percent (30/37) of the farms managed the quarantine unit properly, i.e. using the all-in/all-out principle and having a quarantine duration of minimum 28 days. However, combined, only 10 % (4/39) of the farms complied with the optimal introduction procedures of all three categories, i.e. purchasing policy, quarantine building, and quarantine management.

Table 5. Compliance of the farms to the optimal introduction procedures.

Variable	n	%
Application of the correct procedures regarding the purchasing policy (n = 39)		
Always working with the same origin farm	*38*	*97*
High health status of the origin farm	*31*	*79*
Hygienic requirements for the transport vehicle	*25*	*64*
All of the above	*21*	*54*
Application of the correct procedures regarding the quarantine building (n = 37)		
Quarantine unit with separate air volume	*25*	*68*
Hygiene lock for the quarantine unit	*20*	*54*
All of the above	*14*	*38*
Application of the correct procedures regarding the quarantine management (n = 37)		
Application of the all-in/all-out principle	*32*	*86*
Quarantine period duration of minimum 28 days	*34*	*92*
All of the above	*30*	*81*

DISCUSSION

This study investigated the introduction procedures of breeding gilts in Belgian pig farms and more specifically which purchasing, quarantine, and acclimation practices pig farmers use. Furthermore, current field practices were contrasted with the optimal introduction procedures.

A lot of attention was paid to the wording of the questions and structure of the questionnaire. The questionnaire was pre-tested by colleagues and experts in the field. Despite the fact that the questionnaires were filled in during an on-farm interview, it was for some questions unclear how they were interpreted precisely by the veterinarian and/or the farmer. Regarding the vaccination strategies for instance, it was unclear whether the answers related to the rearing phase and quarantine period only, or whether vaccinations applied during the first gestation of the gilts were included as well. The same was true for the housing of the gilts. Nevertheless, the answers were assumed to be applicable to the rearing and quarantine phase and they were not excluded from the analysis. The absence of the answering option 'sometimes' for some questions is another limitation of the questionnaire design. By only providing the options 'yes' and 'no', it is assumed that farmers always work according to the same principles, which might not necessarily or always be the case.

Pig production in Belgium is mostly located in Flanders, the northern Dutch-speaking part of the country. In 2020, there were approximately 6.1 million pigs in Belgium: 1.6 million piglets (< 20 kg), 4 million fattening pigs (> 20 kg), and 400 000 sows. Ninety-seven percent of the sows were located in Flanders, and only 3 % in Wallonia, the southern French-speaking part of the country (StatBel, 2020). All questionnaires were collected in Flanders. For 2019, the number of herds are known as well. The sows in Belgium were housed on 1678 different farms, of which 90 % (n = 1518) located in Flanders, and 10 % (n = 160) in Wallonia (StatBel, 2019a). In Flanders, 60 % (911/1518) of the sow herds had more than 150 pigs, while in Wallonia, only 16 % (25/160) of the sow herds had more than 150 pigs (StatBel, 2019b). In terms of herd size, the sow herds in the present study were representative for other sow herds in Belgium (Caekebeke et al., 2020; Filippitzi et al., 2018; Postma et al., 2017). Also the batch farrowing systems of the farms in the present study were in line with other studies, showing that the 3- and 4-week batch farrowing system are most commonly used in Belgium (Caekebeke et al., 2020; Postma et al., 2016; Postma et al., 2017).

Garza-Moreno et al. (2017) found that in Europe, replacement gilts were purchased from another farm in 45 % of the cases, whereas own rearing of gilts occurred on 32 % of the farms. On the remaining 23 % of the farms, there was a combination of purchasing and own rearing of gilts. Chantziaras et al. (2018) found similar percentages: 56 % of the farms purchased breeding gilts and 44 % reared own gilts. Caekebeke et al. (2020) found that more than half of the Belgian farms did not purchase any animals. In our study, we found that 57 % (39/68) of the farms purchased breeding gilts and 43 % (29/68) bred their own gilts and no combination of these methods was used. Purchasing breeding gilts is a risk factor for introduction of pathogens into the farm (Pritchard et al., 2005), e.g. porcine reproductive and respiratory syndrome virus (PRRSV) (Firkins & Weigel, 2004). Three percent (1/39) of the farms indicated that the breeding gilts originated from multiple origin farms, which is generally considered as a clear risk. Purchasing breeding gilts from multiple origin farms increases the risk of reinfection of specific pathogen free (SPF) farms with *M. hyopneumoniae*, and can result in a large number of slaughter pigs seropositive for *A. pleuropneumoniae* serovar 2 (Jorsal & Thomsen, 1988; Maes et al., 2001).

Transport vehicles of livestock are found to be an important source of contamination for many pathogens, such as classical swine fever (Fritzemeier et al., 2000), *M. hyopneumoniae* (Hege et al., 2002), *A. pleuropneumoniae* (Hege et al., 2002), and *B. hyodysenteriae* (Windsor & Simmons, 1981). Therefore, transport vehicles should be cleaned and disinfected according to a strict protocol before they are allowed to enter the premises (Dewulf et al., 2018). However, only 28 % (7/25) of the farms indicated cleaning and disinfection as a requirement for the transport truck. In addition, 28 % (7/25) of the farmers only allowed transport vehicles on their farm in the morning when no other farms had been visited or on Monday morning when no other farms had been visited in the weekend. These requirements consider that in these cases, transport vehicles were cleaned, disinfected, and empty for at least 12 hours.

On 5 % (2/39) of the farms, it was indicated that the purchased animals were not isolated in a quarantine unit. One of those farmers specified that the gilts were housed immediately in a compartment where other pigs of the farm were present as well, and this farmer was aware of the risk associated with this procedure. The reason they did not use a quarantine was not asked. Sixty-eight percent (25/37) of the farms had a quarantine unit where the air volume was separated from other pigs on the farm, namely the farms with an external quarantine unit, the farms with an isolated stable, and the farms that indicated that there was a separate ventilation system in the quarantine compartment. Isolation of purchased breeding gilts can reduce the risk of pathogen introduction in the herd (Firkins & Weigel, 2004). In the North American swine industry it is common to house breeding gilts in a specialized gilt development unit (GDU). GDUs are used to raise gilts and to gradually adapt them to the health status of the sow herd (Garza-Moreno et al., 2018).

Pathogens can be transmitted indirectly through contaminated hands, clothing, and boots (Filippitzi et al., 2018). Therefore, it is important to have a separate hygiene lock for the quarantine unit, with water supply for hand hygiene and specific clothing and boots for the quarantine unit. Only 54 % (20/37) of the farms had a separate hygiene lock for the quarantine and it is not known if hygiene measures were performed well according to a strict protocol. Pathogen transmission between gilts in the quarantine unit and other animals on the farm can occur when no measures are taken in between entering the quarantine unit and the compartments of the other animals (Pritchard et al., 2005). Risk of pathogen transmission depends on whether people visit the quarantine unit as the last task of the working day, and if they start the next day with clean clothing and cleaned and disinfected boots. This is especially important in the initial phase of the quarantine period, when the main goal is avoiding possible introduction of pathogens by newly purchased animals into the farm (Dewulf et al., 2018). The frequency and duration of visits to the quarantine unit by the farmer and/or employees could have an influence on this as well. Moreover, this information is important to estimate whether monitoring for clinical signs in the breeding gilts was performed properly. Nevertheless, information on frequency, duration, and time point of visits to the quarantine unit was not included in the questionnaire.

Pritchard et al. (2005) suggested that the quarantine period should last at least three to four weeks. According to Neumann and Hall (2019) the duration of the quarantine period typically varies between 30 and 60 days. Both studies agree that the duration of the quarantine period depends on the specific pathogens of concern. In Belgian pig production, there is limited legislation on the application and duration of a quarantine period. Article 7 of the Belgian Royal Decree of 18 June 2014, regarding measures to prevent notifiable swine diseases, states that farms are the first four weeks after purchasing of new animals only allowed to transport fattening pigs to the slaughterhouse. However, if a quarantine period of four weeks is applied for the newly purchased animals, the farms are allowed to send piglets to other farms, or to transport sows that will be culled to the slaughterhouse (Royal Decree, 2014). Since the threat of African Swine Fever (ASF) in Belgium, extra legal requirements were in place, which are listed in the Ministerial Decision of 26 September 2018 regarding urgent measures to control ASF. Chapter 2 of this decree includes biosecurity measures for the entire country, e.g. all pigs that enter a herd must be housed separately for four weeks (Article 15), and group treatment of clinically sick animals is only allowed after a negative ASF-diagnosis is confirmed by laboratory analysis (Article 16) (Ministerial Decision, 2018). The median duration of the quarantine period in this study was 42 days, which proves that 50 % of the farms had a quarantine period longer than or equal to six weeks, which should be long enough to monitor clinical signs of several diseases and to perform laboratory testing. However, previous studies have shown that monitoring for diseases during the quarantine period is not very common. In the study of Garza-Moreno et al. (2017), 28 % of the farms performed diagnostics for *M. hyopneumoniae* in the purchased breeding gilts. Lambert et al. (2012) found that 11 % of the farms evaluated the PRRSV-status of the gilts at the end of the quarantine period. In our study, only 16 % (11/68) of the farmers monitored breeding gilts for presence of *B. hyodysenteriae* (via PCR) and/or other pathogens (via ELISA). Furthermore, interpretation of serological data requires knowledge on vaccination of the pigs (vaccination yes/no, product used, scheme), since most serological tests make no distinction between antibodies originating from infection or vaccination (Maes et al., 2018). To this end, it is important to know if the purchased animals already received vaccinations at the origin farm. For all pathogens, vaccination was done at the origin farm, except for the pathogens related to intestinal health, i.e. porcine rotavirus type A, *Escherichia coli*, and

Clostridium spp. A possible explanation could be that breeding gilts only receive those vaccinations at the end of first gestation, in order to provide their offspring with lactogenic immunity. Other techniques can also be used to detect the pathogen, antigens, or genetic material in blood, tonsil samples, nasal samples, laryngeal and tracheal swabs, fluid of bronchoalveolar lavage, and feces (Maes et al., 2018). Assessing the health status of the breeding gilts by clinical evaluation and laboratory testing, provides valuable information to the farmers, as it could help them to prevent pathogen introduction and/or maintain a free-status for specific pathogens, for example *B. hyodysenteriae*. In total, 79 % (31/39) of the farmers was aware of the health status of the origin farm, hence they knew for which pathogens these farms were free. Purchasing gilts from an SPF farm could also be useful to keep the farms free of specific diseases. This can be considered as primary disease prevention, since the main goal is to avoid introduction of specific pathogens in a farm (Toma et al., 1999). The number of SPF farms in Belgium is not known, in contrast to Denmark where 2300 sow herds (80 % of the Danish sows) have an SPF health status (TS Hansen, personal communication).

Vaccination of purchased breeding gilts was practiced in all farms and was therefore the most commonly used acclimation practice. A variety of vaccination strategies were used and on all farms, breeding gilts were vaccinated against at least two pathogens. This is in accordance to Garza-Moreno et al. (2017), who found that vaccination was the most important gilt acclimation practice for *M. hyopneumoniae* in Europe. Most vaccines do not give a full protection, do not prevent infection, and cannot eliminate pathogens from a herd. Nevertheless, vaccination is very important because it reduces the risk of pathogen transmission, clinical signs, lesions, and performance losses due to disease (Maes et al., 2018). Subsequently, vaccination may be cost-efficient, even in subclinically infected herds (Maes et al., 2003). Pieters and Fano (2016) suggested a method of strategic exposure of gilts to *M. hyopneumoniae* by vaccination at a young age, aiming to let them undergo the infectious process, and recover and gain immunity. In this way, these animals do not shed *M. hyopneumoniae* anymore when they are introduced in the sow herd. For some pathogens, e.g. *B. hyodysenteriae* and *S. suis*, no commercial vaccines are available, and sometimes autogenous vaccines are used. However, little is known on the efficacy and safety of these vaccines (Haesebrouck et al., 2004). In this study, two farmers used autogenous vaccines.

Bringing feces from piglets in the rearing or quarantine unit was sometimes used as an acclimation practice. Interpreting Article 36 and 39 of the Regulation (EC) No 1069/2009 regarding animal by-products, feces could be used as a derived product for disease prevention by controlled exposure if advised by the veterinarian. However, feces cannot be used as a source of nutrition for animals and pig feed and drinking water should be kept clean and safe at all time (European Council Regulation, 2009). It is better not to use alternative acclimation practices, such as contact with feces or placenta tissue, since it is not known which pathogens are in and thus it is unclear which pathogens are spread. Therefore, it is in contrast with internal biosecurity principles, aiming to prevent or limit pathogen transmission within the herd (Dewulf et al., 2018).

On 82 % (56/68) of the farms, the breeding gilts were housed in groups. On 8 % (4/53) of those farms, the stocking density of the breeding gilts was less than 1 m². This is not in accordance with the minimal legal requirements, which state that the surface area of pigs weighing more than 110 kg should be 1 m²/pig, and preferably even higher (Dewulf et al., 2007). Moreover, higher stocking density can lead to a higher level of disease and can predispose to leg weaknesses and claw disorders (Jørgensen, 2003; Pointon et al., 1985).

If the separate categories of the introduction procedures are considered, namely purchasing policy, quarantine building, and quarantine management, 54 % (21/39), 38 % (14/37), and 81 % (30/37) of the farms respectively, complied with the optimal procedures. However, only 10 % (4/39) of the farms complied with all optimal introduction procedures. This indicated that there is a lot of room for improvement and efforts should be made to raise the farmers' awareness. Herd veterinarians could play a key role in improving biosecurity related to the introduction procedures of breeding gilts in pig herds. This could be facilitated by the use of a checklist or a step-wise protocol for the purchasing of breeding gilts.

CONCLUSION

Fifty-seven percent (39/68) of the farms purchased breeding gilts and there was a lot of variation in the frequency of purchase and the age at which gilts are purchased. On 95 % (37/39) of those farms, a quarantine unit was used, where on most farms the quarantine was located on the farm itself. In general, the gilts were kept in quarantine for six weeks. Vaccination was the most commonly applied acclimation practice, although in some farms exposure of gilts to farm-specific microorganisms was done by providing feces of suckling piglets and bringing the gilts in contact with sows before culling. Only 10 % (4/39) of the farms applied the optimal introduction procedures of breeding animals, so there is a lot of room for improvement and farmers' awareness on this topic should be raised.

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CHAPTER 4 | OPTIMIZING INTERNAL BIOSECURITY ON PIG FARMS BY ASSESSING MOVEMENTS OF FARM STAFF

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Adapted from

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ABSTRACT

For internal biosecurity, it is important to separate different age groups in a pig farm and to stick to specific working lines when visiting the barns. Currently, there is no research on the movements of farm staff on pig farms. The objectives of this observational study were to assess movements of farm staff on pig farms, to assess risky movements and to investigate whether movements differ according to time (week of the batch farrowing system (BFS) and weekday vs. weekend) and unit (farrowing, gestation/insemination, nursery, and fattening unit). Five commercial sow farms participated and on each farm, an internal movement monitoring system was installed. Detection points were installed throughout the farm and workers had to wear a personal beacon. Movement data were collected from 1 December 2019 until 30 November 2020. The following sequence of movements was considered as safe: (1) hygiene lock, (2) farrowing, (3) gestation/insemination, (4) nursery, (5) fattening, (6) quarantine unit, and (7) cadaver storage. Movements in the opposite direction were considered as a risk, unless a hygiene lock was visited in between. The total number of movements differed according to week of the BFS and was highest in insemination and farrowing week. The percentage of risky movements was influenced by week of the BFS for two farms and was highest around weaning. The percentage of risky movements varied between farms and ranged from 9 to 38 %. There were more movements on a weekday compared to a weekend day. There were more movements towards the farrowing and gestation/insemination unit in insemination and farrowing week compared to other weeks of the BFS, but week of the BFS had no impact on movements towards nursery and fattening unit. This study showed that there were a lot of (risky) movements on pig farms and that these movements varied according to week of the BFS, day of the week, and unit. This study creates awareness, which could be a first step in optimizing working lines. Future research should focus on why certain risky movements occur and how these can be avoided to achieve better biosecurity and higher health status on farms.

INTRODUCTION

Infections with specific pathogens commonly occur in pig farms and may result in major economic losses for the farmer. Such pathogens are transmitted through different routes, either directly via contact with infected animals or indirectly via people, semen, manure, rodents, aerosol, feed, water, or fomites (Filippitzi et al., 2018). Biosecurity measures on a farm aim to limit or even prevent the transmission of pathogens. All measures aiming to reduce the risk of pathogen introduction on a farm are grouped as external biosecurity measures, while those aiming to reduce the spread of pathogens within a farm are grouped as internal biosecurity measures. The implementation of biosecurity measures has multiple benefits, such as a reduced disease incidence and less antimicrobial use (Collineau et al., 2017), improved production (Laanen et al., 2013; Postma et al., 2016), and improved farm profitability (Collineau et al., 2017; Rojo-Gimeno et al., 2016). A previous study in France has shown that farm structure and working lines were significantly associated with a lower antimicrobial use (Lannou et al., 2012).

The European Animal Health Law emphasizes the importance of biosecurity to prevent the spread of infectious diseases to and within farms. Farm staff should acquire the appropriate knowledge and they should take action to minimize the spread of pathogens by working according to the correct working lines (European Council Regulation, 2016). Each visit to a pig farm from both farm staff and visitors should start in a hygiene lock, where farm-specific clothing and footwear can be put on and where hands can be properly washed (Dewulf et al., 2018; Neumann & Hall, 2019). Additional hygiene locks for each animal category could further reduce the risk of pathogen transmission (Dewulf et al., 2018). Furthermore, farm staff should follow a specific sequence in visiting the units with different animal categories. Younger animals are more susceptible to various pathogens due to decreased maternal immunity while they have not yet developed a mature active immunity, whereas older animals are considered to be more robust but at the same time they may also harbor more infectious agents due to previous infections. Often these will remain unnoticed as a result of subclinical infection status. Therefore, movements or daily work should ideally be performed from young to old and from healthy to sick animals, thus according to the following sequence: (1) hygiene lock, (2) farrowing unit, (3) gestation/insemination unit, (4) nursery unit, (5) fattening unit, (6) quarantine unit, and (7) cadaver storage (Dewulf et al., 2018; Vangroenweghe et al., 2009). Movements in the opposite direction are considered risky as they may cause pathogen transmission. Therefore, biosecurity measures aim at separating different age groups as much as possible. If these (virtual) separations are breached in specific units or on specific time points, then the overall biosecurity goes down and the efforts made in the other units or on different time points may be nullified.

A way to increase the awareness and motivation of pig farmers is to evaluate the biosecurity in pig farms (Alarcón et al., 2021). The most common way to evaluate biosecurity is an assessment based on scores, such as Biocheck.UGent (Biocheck.UGent, 2023). Although these scoring systems are good for creating awareness, they do not evaluate every component of the biosecurity in detail as this would make the overall assessment too complex and laborious. In the Biocheck.UGent survey for pigs, there are two questions related to movements of farm staff, namely: (1) Are diseased pigs consistently handled/visited after the healthy ones?, and (2) Is all the farm work performed from younger pigs to older pigs (Biocheck.UGent, 2023)? To verify if these conditions were applied consistently, Precision Livestock Farming (PLF) could be used. PLF is a concept in animal production where modern technologies, such as sensors and algorithms, are used to automatically gather data about the animals in order to optimize management practices (Neethirajan, 2017; Piñeiro et al., 2019). Some examples in pig production are electronic feeders to register feed intake by the animals (Koketsu et al., 1996), sensors that register estrus behavior in sows to optimize the moment of insemination (Labrecque & Rivest, 2018), real-time sound analysis for health monitoring (Berckmans et al., 2015), and sensors monitoring the stable climate 24/7

(Healthy Climate Solutions, 2023). Another example is the real-time internal movement monitoring system Biorisk® developed by Animal Data Analytics. This sensor is not for the animals, but for farm staff. The system can be used to monitor the working lines of farm staff in pig farms and to preserve good biosecurity routines (Animal Data Analytics, 2023).

Currently, there is no information on the movements of farm staff in pig farms. The general objective of the present study was to investigate the movements of staff in commercial pig farms. Possible movement differences according to time (week of a batch farrowing system (BFS) and day of the week) and unit (farrowing, gestation/insemination, nursery and fattening unit) in the farm were also investigated.

MATERIALS AND METHODS

STUDY DESIGN

This observational study was performed from 1 December 2019 until 30 November 2020. Farms were selected based on willingness to participate and to install wireless internet connection in all barns. Five commercial sow farms participated in the study and their characteristics are described in Table 1. In a BFS, the main tasks on a farm, i.e. weaning, insemination, and farrowing, are divided over different weeks. For farm A, working in a 3-week system, the following terminology for the different weeks will be used: weaning (the week in which a group of sows is weaned), insemination (the week in which a group of sows is inseminated), and farrowing (the week in which a group of sows farrows). For farms B, C, D, and E, working in a 4-week system, the following terminology will be used: weaning (the week in which a group of sows is weaned), insemination + farrowing (the week in which a group of sows is inseminated, and another group of sows is planned to farrow), 'no main activities 1' (the week after farrowing, when suckling piglets are handled in the farrowing unit, e.g. iron injection), and 'no main activities 2' (the week in which the nursery pigs are moved to the fattening unit).

For the housing of the animals, the terms room, unit, and barn will be used. Room refers to a room where animals of the same category are present, except for the farrowing room in which both sows and suckling piglets are present. A unit can consist of one or more rooms; and a barn can consist of one or more units. In some cases, a barn consists of different types of units, e.g. a barn with a farrowing and a gestation/insemination unit.

In all five farms, the internal movement monitoring system Biorisk® developed by Animal Data Analytics was installed (Animal Data Analytics, 2023). For this purpose, detection points were installed in the rooms of the different animal categories on the farm. The detection points had a range of eight meters and were installed in such a way to cover all rooms and entrances of the farm. In some units, only one detection point was needed, while in others more than one was needed to ensure all rooms were covered. A time filter was set for each detection point to avoid wrong detections in places where a detection point was too close to a corridor. All farm workers had to wear a small personal Bluetooth® transmitter, called a beacon. This transmitter sent a signal to the detection points, allowing the detection of the movements of farm staff. A wireless internet connection was needed to send data from the detection points to an online platform for further analysis (Figure 1). The number of detection points and farm workers contributed to the total number of movements on a farm.

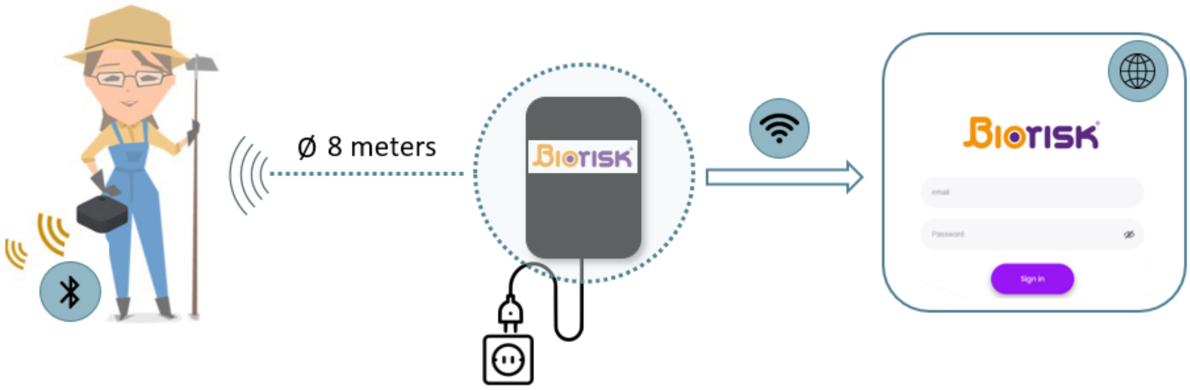

Figure 1. Graphical representation of the Biorisk® system developed by Animal Data Analytics.

The biosecurity status of the farms was determined using the risk-based biosecurity quantification tool Biocheck.UGent. This tool enables an objective quantification of the biosecurity status of the farm. Based on a questionnaire, a score between 0 and 100 % is given in different categories. Zero means a lack of any biosecurity measures, while 100 means perfect biosecurity (Biocheck.UGent, 2023). The overall biosecurity scores and the subtotal for the external and internal biosecurity are shown in Table 1.

Table 1. Characteristics of the five farms that participated in the study.

	Farm A	Farm B	Farm C	Farm D	Farm E
Type of farm	Farrow-to-finish	Farrow-to-finish	Farrow-to-finish	Farrow-to-wean	Farrow-to-finish
Batch farrowing system (… week system)	3	4	4	4	4
Number of full-time employees	1	1	2	5	3
Sow breed	PIC	TN70	Hypor	Danbred	Danbred
Number of animals					
Sows	280	480	300	780	600
Nursery pigs	600	2000	1450	2500	3600
Fattening pigs	2500	1200	480	0	1000
Liveborn piglets per litter	13.1	14.6	14.1	17.5	16.6
Pre-weaning mortality (%)	12.0	13.4	13.7	17.0	7.9
Number of rooms (number of detection points)					
Hygiene lock	1 (1)	1 (1)	2 (2)	2 (2)	1 (1)
Farrowing unit	8 (3)	10 (10)	3 (3)	7 (3)	1 (2)
Gestation/insemination unit	2 (2)	7 (6)	3 (4)	8 (8)	6 (7)
Nursery unit	8 (3)	6 (1)	20 (4)	12 (3)	7 (5)
Fattening unit	11 (11)	6 (3)	6 (1)	0 (0)	5 (3)
Quarantine unit	2 (1)	1 (1)	1 (1)	2 (2)	4 (1)
Cadaver storage	1 (1)	1 (1)	1 (0)	1 (1)	1 (1)
Shower in the hygiene lock	No	No	No	Yes	Yes
Separate clothing and footwear for different units	No	No	No	Yes	Yes
Measures needed to enter the quarantine unit	Boots in disinfection bath	None	Changing boots	Changing coverall and boots	Changing coverall and boots
Location of cadaver storage	Close to the barns	Near to public road (far from the barns)	Near to public road (far from the barns)	Near to public road (far from the barns)	Near to public road (far from the barns)
Biosecurity scores (%)					
Total	60	67	81	72	86
External	66	76	87	77	83
Internal	54	57	74	66	88

Movements from the farrowing unit to the gestation/insemination unit, followed by the nursery unit, fattening unit and finally, the quarantine unit and cadaver storage, were considered as safe movements (Dewulf et al., 2018). Movements in the opposite direction were considered as risk, unless a hygiene lock was visited in between (Figure 2). Movements between rooms of the same type, e.g. farrowing to farrowing room, were also considered as safe movements, except for a movement from a quarantine to another quarantine room. In total, 49 different movements could be distinguished, 33 of them were considered as safe and 16 as risky. Farm C did not have a power socket available at the cadaver storage. Therefore, in this farm, movements from and to the cadaver storage could not be taken into consideration.

From: / **To**:	Hygiene lock	Farrowing	Gestation/ insemination	Nursery	Fattening	Quarantine	Cadaver
Hygiene lock	safe	safe	safe	safe	safe	safe	safe
Farrowing	safe	safe	safe	safe	safe	safe	safe
Gestation/ insemination	safe	risky	safe	safe	safe	safe	safe
Nursery	safe	risky	risky	safe	safe	safe	safe
Fattening	safe	risky	risky	risky	safe	safe	safe
Quarantine	safe	risky	risky	risky	risky	risky	safe
Cadaver	safe	risky	risky	risky	risky	risky	safe

Figure 2. Definition of safe and risky movements by persons in the five farms included in the study.

DATA ANALYSIS

Descriptive statistics were performed for the continuous variables. Normality distribution was analyzed graphically via histograms and Q-Q plots. All movement data were not normally distributed; therefore, the median, minimum and maximum values were used.

Since the number of detection points and the number of animals varied between the farms, we standardized the total number of daily movements to allow comparison between the farms. For movements towards the farrowing and the gestation/insemination unit, the movements were standardized per detection point and per 100 sows. For movements towards the nursery and fattening unit, the movements were standardized per detection point and per 1000 nursery or fattening pigs.

A non-parametric independent samples Kruskal-Wallis test with Bonferroni correction was used to analyze potential differences in the total number of daily movements, percentage of risky movements, movements towards specific units within the farm, and number of movements towards specific units standardized by farm size between the different weeks of the BFS. For the latter, only the farms working in a 4-week BFS (B – E) were included in the analysis. Farm D was not included in the analysis of the movements towards the fattening unit, since there were no fattening pigs present on this farm. A non-parametric independent-samples Mann-Whitney U test was used to analyze potential differences in the total number of movements and percentage of risky movements between a weekday (Monday to Friday) vs. a day on the weekend (Saturday and Sunday). *P*-values below 0.05 were considered to be statistically significant. All statistical analyses were performed using IBM® SPSS® Statistics for Windows Version 28 (IBM Corp., Armonk, N.Y., USA).

RESULTS

MOVEMENTS OF FARM STAFF – BIOCHECK.UGENT AND FARM SIZE

The Biocheck.UGent survey for pigs was carried out in all farms. Specific attention was paid to the two questions related to movements of farm staff. Farms C and E stated that diseased pigs were consistently handled/visited after the healthy ones. Farms A, C, D, and E stated that all the farm work was performed from younger to older pigs.

The overall median percentages of risky movements on the farms were 11 %, 33 %, 36 %, 15 %, and 14 %, and the numbers of sows were 280, 480, 300, 780, and 600 for farms A, B, C, D, and E, respectively. The lowest percentage of risky movements was seen on the smallest farm (farm A). There were a few more risky movements on the largest farms (farms D and E), and the highest percentage of risky movements was seen on the medium-sized farms (farms B and C).

MOVEMENTS OF FARM STAFF ACCORDING TO THE WEEK OF THE BATCH FARROWING SYSTEM

Table 2 shows the total number of daily movements and the percentage of risky movements in the different weeks of the BFS. The total number of movements significantly differed according to the week of the BFS for farms B ($p = 0.005$), C ($p < 0.001$), D ($p < 0.001$), and E ($p = 0.029$), with the highest number of total movements during insemination and farrowing week, followed by weaning week. The percentage of risky movements significantly differed according to the week of the BFS for farms C ($p = 0.014$) and D ($p = 0.020$), with the highest percentages in the weaning week.

Table 2. The median (min. – max.) number of daily movements and the percentage of risky movements in the different weeks of the batch farrowing system in farm A, working in a 3-week system, and farms B, C, D, and E, working in a 4-week system.

	Total number of daily movements (*n*)		Percentage of risky movements (%)	
Farm A				
Weaning	32^{a}	(2 – 76)	11^{a}	(0 – 30)
Insemination	32^{a}	(9 – 75)	9^{a}	(0 – 32)
Farrowing	33^{a}	(13 – 90)	11^{a}	(0 – 35)
Farm B				
Weaning	32^{ab}	(2 – 633)	33^{a}	(0 – 52)
Insemination + farrowing	53^{a}	(2 – 348)	33^{a}	(0 – 46)
No main activities 1	33^{b}	(1 – 349)	33^{a}	(0 – 45)
No main activities 2	38^{b}	(2 – 320)	32^{a}	(0 – 50)
Farm C				
Weaning	58^{a}	(21 – 244)	38^{a}	(10 – 46)
Insemination + farrowing	64^{a}	(14 – 236)	35^{b}	(17 – 47)
No main activities 1	49^{b}	(14 – 166)	36^{ab}	(20 – 44)
No main activities 2	41^{c}	(13 – 103)	36^{ab}	(8 – 50)
Farm D				
Weaning	71^{ab}	(10 – 247)	16^{a}	(6 – 27)
Insemination + farrowing	85^{a}	(8 – 210)	14^{ab}	(4 – 27)
No main activities 1	66^{bc}	(2 – 173)	15^{ab}	(0 – 32)
No main activities 2	52^{c}	(7 – 273)	13^{b}	(0 – 28)
Farm E				
Weaning	43^{ab}	(5 – 487)	14^{a}	(0 – 44)
Insemination + farrowing	57^{a}	(2 – 351)	10^{a}	(0 – 60)
No main activities 1	31^{ab}	(1 – 629)	12^{a}	(0 – 40)
No main activities 2	35^{b}	(1 – 544)	14^{a}	(0 – 40)

^{abc} Within each farm and within a column, values with different superscript differed significantly ($p < 0.05$)

MOVEMENTS OF FARM STAFF DURING WEEKDAYS VS. DAYS ON THE WEEKEND

Possible differences in movements between a weekday vs. a day on the weekend were investigated (Table 3). On all farms, there was a higher total number of movements on a weekday than on a weekend day. This difference was statistically significant for all farms, except for farm C. On farms B, C, and D, there was a significant difference in the percentage of risky movements on a weekday than on a weekend day. On farms B and D there were less risky movements during the weekend, while on farm C there were more risky movements during the weekend.

Table 3. The median (min. – max.) number of daily movements and the percentage of risky movements on a weekday and a weekend day for the different farms (n = 5).

	Total number of daily movements (n)			Percentage of risky movements (%)		
Farm	Week	Weekend	p-value	Week	Weekend	p-value
A	37 (2 – 90)	24 (10 – 51)	< 0.001*	10 (0 – 32)	11 (0 – 35)	0.216
B	45 (2 – 633)	27 (1 – 349)	< 0.001*	33 (0 – 50)	31 (0 – 52)	0.046*
C	52 (14 – 236)	51 (13 – 244)	0.242	36 (10 – 50)	37 (8 – 44)	0.041*
D	80 (10 – 273)	34 (2 – 125)	< 0.001*	15 (0 – 29)	13 (0 – 32)	0.003*
E	50 (1 – 629)	31 (1 – 544)	0.001*	13 (0 – 60)	11 (0 – 60)	0.661

* P-values below 0.05 were considered statistically significant

MOVEMENTS OF FARM STAFF TOWARDS FARROWING AND GESTATION/INSEMINATION UNIT

Table 4 shows the total number of movements and the percentage of risky movements towards the farrowing and gestation/insemination unit. The total number of movements and the percentage of risky movements towards the farrowing unit significantly differed for the different weeks of the BFS for all the farms (p < 0.05), except for the risky movements on farm A (p = 0.403) and farm E (p = 0.259). There were more movements on a day in the insemination and farrowing week. The percentage of risky movements was highest during the weaning week.

The total number of movements and the percentage of risky movements towards the gestation/insemination unit significantly differed for the different weeks of the BFS for all the farms (p < 0.05), except for farm A. Farm workers had more movements towards the gestation/insemination unit in insemination and farrowing week. For the percentage of risky movements towards the gestation/insemination unit, there was not one specific week of the 4-week system with more risky movements and there was some variation between the farms (Table 4).

Table 4. Median (min. – max.) number of daily movements (total movements and percentage of risky movements) towards the farrowing unit and the gestation/insemination unit in the different weeks of the batch farrowing system in farm A, working in a 3-week system, and farms B, C, D, and E, working in a 4-week system.

	Total number of movements (*n*)		Percentage of risky movements (%)	
	Farrowing	Gestation/ insemination	Farrowing	Gestation/ insemination
Farm A				
Weaning week	4^{a} (1 – 12)	4^{a} (1 – 17)	40^{a} (0 – 100)	33^{b} (0 – 100)
Insemination week	2^{b} (1 – 8)	5^{a} (1 – 16)	33^{a} (0 – 100)	16^{a} (0 – 100)
Farrowing week	4^{a} (1 – 11)	4^{a} (1 – 11)	33^{a} (0 – 100)	20^{a} (0 – 100)
Farm B				
Weaning	6^{b} (1 – 158)	16^{ab} (1 – 344)	100^{a} (0 – 100)	20^{b} (0 – 100)
Insemination + farrowing	15^{a} (1 – 169)	25^{a} (2 – 105)	94^{b} (0 – 100)	13^{a} (0 – 45)
No main activities 1	7^{b} (1 – 94)	14^{b} (1 – 169)	94^{b} (0 – 100)	21^{b} (0 – 100)
No main activities 2	8^{b} (1 – 36)	16^{b} (1 – 151)	100^{b} (0 – 100)	21^{b} (0 – 100)
Farm C				
Weaning	22^{a} (6 – 95)	19^{a} (3 – 103)	89^{a} (33 – 100)	6^{ab} (0 – 57)
Insemination + farrowing	23^{a} (3 – 94)	23^{a} (1 – 123)	88^{a} (50 – 100)	8^{a} (0 – 100)
No main activities 1	18^{ab} (4 – 70)	14^{b} (4 – 57)	83^{b} (50 – 100)	4^{b} (0 – 50)
No main activities 2	16^{b} (4 – 43)	14^{b} (2 – 37)	87^{ab} (25 – 100)	0^{b} (0 – 38)
Farm D				
Weaning	22^{a} (1 – 120)	28^{a} (4 – 97)	40^{a} (13 – 100)	8^{ab} (0 – 24)
Insemination + farrowing	39^{b} (3 – 86)	24^{ab} (1 – 77)	33^{b} (8 – 60)	6^{a} (0 – 29)
No main activities 1	27^{a} (2 – 87)	19^{bc} (1 – 78)	35^{b} (3 – 67)	7^{a} (0 – 30)
No main activities 2	13^{c} (2 – 34)	16^{c} (2 – 39)	36^{b} (0 – 63)	11^{b} (0 – 50)
Farm E				
Weaning	24^{a} (1 – 106)	8^{a} (1 – 49)	67^{a} (3 – 100)	33^{a} (0 – 100)
Insemination + farrowing	44^{b} (2 – 129)	7^{ab} (1 – 46)	45^{a} (0 – 100)	9^{ab} (0 – 100)
No main activities 1	24^{ab} (1 – 120)	5^{b} (1 – 48)	55^{a} (0 – 100)	5^{b} (0 – 100)
No main activities 2	19^{a} (1 – 147)	6^{ab} (1 – 46)	55^{a} (0 – 100)	0^{b} (0 – 100)

[abc] Within each farm and within a column, values with different superscript differed significantly ($p < 0.05$)

MOVEMENTS OF FARM STAFF TOWARDS NURSERY AND FATTENING UNIT

Table 5 shows the total number of movements and the percentage of risky movements towards the nursery and fattening unit. Since farm D was farrow-to-wean, there were no movements towards the fattening unit. Regarding the movements towards the nursery unit, we found significant differences in the total number of movements in farm C ($p < 0.001$) and the percentage of risky movements in farm E ($p < 0.001$). The movements towards the fattening unit did not significantly differ between the weeks of the BFS.

Table 5. Median (min. – max.) number of daily movements (total movements and percentage of risky movements) towards the nursery unit and fattening unit in the different weeks of the batch farrowing system in farm A, working in a 3-week system, and farms B, C, D, and E, working in a 4-week system.

	Total number of movements (*n*)		Percentage of risky movements (%)	
	Nursery	Fattening	Nursery	Fattening
Farm A				
Weaning week	2^{a} (1 – 9)	8^{a} (1 – 29)	0^{a} (0 – 100)	5^{a} (0 – 50)
Insemination week	2^{a} (1 – 7)	7^{a} (2 – 26)	0^{a} (0 – 100)	13^{a} (0 – 50)
Farrowing week	2^{a} (1 – 9)	7^{a} (2 – 33)	0^{a} (0 – 100)	7^{a} (0 – 33)
Farm B				
Weaning	2^{a} (1 – 11)	3^{a} (1 – 19)	0^{a} (0 – 100)	0^{a} (0 – 50)
Insemination + farrowing	1^{a} (1 – 8)	3^{a} (1 – 16)	0^{a} (0 – 100)	0^{a} (0 – 100)
No main activities 1	2^{a} (1 – 13)	3^{a} (1 – 13)	0^{a} (0 – 100)	0^{a} (0 – 50)
No main activities 2	1^{a} (1 – 19)	3^{a} (1 – 38)	0^{a} (0 – 100)	0^{a} (0 – 67)
Farm C				
Weaning	6^{ab} (1 – 30)	2^{a} (1 – 24)	0^{a} (0 – 77)	0^{a} (0 – 50)
Insemination + farrowing	6^{b} (1 – 19)	2^{a} (1 – 6)	0^{a} (0 – 40)	0^{a} (0 – 50)
No main activities 1	5^{ac} (1 – 16)	2^{a} (1 – 7)	0^{a} (0 – 33)	0^{a} (0 – 0)
No main activities 2	4^{c} (1 – 13)	2^{a} (1 – 6)	0^{a} (0 – 83)	0^{a} (0 – 100)
Farm D				
Weaning	7^{a} (1 – 47)	-	0^{a} (0 – 10)	-
Insemination + farrowing	6^{a} (1 – 33)	-	0^{a} (0 – 13)	-
No main activities 1	6^{a} (1 – 31)	-	0^{a} (0 – 0)	-
No main activities 2	8^{a} (1 – 134)	-	0^{a} (0 – 0)	-
Farm E				
Weaning	12^{a} (1 – 146)	3^{a} (1 – 23)	0^{a} (0 – 100)	0^{a} (0 – 100)
Insemination + farrowing	15^{a} (1 – 74)	3^{a} (1 – 9)	0^{a} (0 – 100)	0^{a} (0 – 100)
No main activities 1	9^{a} (1 – 269)	3^{a} (1 – 19)	0^{a} (0 – 100)	0^{a} (0 – 56)
No main activities 2	9^{a} (1 – 119)	3^{a} (1 – 17)	9^{b} (0 – 100)	0^{a} (0 – 100)

^{abc} Within each farm and within a column, values with different superscript differed significantly ($p < 0.05$)

NUMBER OF MOVEMENTS TOWARDS THE DIFFERENT UNITS STANDARDIZED BY FARM SIZE

In order to enable proper comparison between farms, the number of movements towards the different units were standardized by number of detection points and farm size. There was a significant effect of the week of the BFS on movements towards the farrowing unit ($p < 0.001$) and gestation/insemination unit ($p < 0.001$) (Table 6). Overall, most movements towards the farrowing unit were made in insemination and farrowing week and the least movements in 'no main activities 2', i.e. the week where nursery pigs are moved to the fattening unit. There were more movements towards the gestation/insemination unit in weaning or insemination and farrowing week compared to the weeks with no main activities. There was no significant effect of the week of the BFS on movements towards the nursery or fattening unit.

Table 6. Median (min. – max.) number of daily movements towards the different units standardized per detection point and per 100 sows for movements towards the farrowing and gestation/insemination unit and per 1000 nursery/fattening pigs for movements towards the nursery and fattening unit in the different weeks of the batch farrowing system in farms B, C, D, and E, working in a 4-week system.

	Movements per detection point per 100 sows (*n*)		Movements per detection point per 1000 nursery/fattening pigs (*n*)	
	Farrowing	Gestation/ insemination	Nursery	Fattening
Farms B, C, D, E				
Weaning	1.1^{a} (0.0 – 10.6)	0.6^{a} (0.0 – 11.9)	0.9^{a} (0.1 – 8.1)	1.1^{a} (0.3 – 12.5)
Insemination + farrowing	1.6^{b} (0.0 – 10.8)	0.6^{a} (0.0 – 10.3)	0.9^{a} (0.1 – 4.4)	1.1^{a} (0.3 – 12.5)
No main activities 1	1.1^{ac} (0.0 – 10.0)	0.4^{b} (0.0 – 5.9)	0.8^{a} (0.1 – 14.9)	1.1^{a} (0.3 – 14.6)
No main activities 2	0.8^{c} (0.0 – 12.3)	0.4^{b} (0.0 – 5.2)	0.7^{a} (0.1 – 17.9)	1.1^{a} (0.3 – 12.5)

[abc] Within a column, values with different superscript differed significantly ($p < 0.05$)

DISCUSSION

The present study elucidated differences in movements of farm staff according to week of the BFS and weekday vs. weekend; and unit, namely towards farrowing, gestation/insemination, nursery and fattening unit. The following movements differed according to the week of the BFS: total number of daily movements (highest in insemination and farrowing week) and percentage of risky movements (highest in weaning week). There were more farm staff movements during a weekday, but the percentage of risky movements was for some farms higher and for others lower on a weekend day. The present study also gained more insight into movements towards the different units. There were more movements of farm staff towards the farrowing and gestation/insemination unit during insemination and farrowing week, compared to other weeks of the BFS. Movements towards the nursery and fattening unit did not differ according to the week of the BFS, except for the total number of movements towards the nursery in one farm and the percentage of risky movements towards the nursery unit in another farm.

According to the results of the Biocheck.UGent survey, all farms except for farm B claimed to organize their work consistently starting with the young animals and then continuing the work in the older animals. However, the results of the present study did not confirm this, as a high percentage of risky movements was observed on the farms. This illustrates that monitoring the behavior of farmers is key to obtain accurate data, as farmers might not always provide the correct answer in observational studies. On larger farms, farmers should implement more biosecurity measures compared to smaller farms, because a larger number of animals also means that more animals can get sick and spread infections. Moreover, larger herds have more contact with the outside world, e.g. by purchasing animals and livestock transport, increasing the risk of infection (Dewulf, 2018; Dewulf & Van Immerseel, 2018). In the present study, there were less risky movements on larger farms compared to medium-sized farms. On these large farms, there were more employees and it is possible that certain employees were only responsible for the work in certain units, resulting in less movements between the different units. Furthermore, previous studies in both pig (Boklund et al., 2004; Boklund et al., 2003; Laanen et al., 2013) and cattle production (Hoe & Ruegg, 2006; Nöremark et al., 2010) have shown that biosecurity measures are better implemented in larger farms.

The total number of daily movements significantly differed according to the week of the BFS for farms B, C, D, and E, all working in a 4-week system. On farm A, working in a 3-week system, there was no difference in the total number of daily movements according to the week of the BFS. A possible explanation could be that the main activities on the farm, i.e. weaning, insemination, and farrowing, are more evenly spread in a 3-week system, leading to a more even distribution of the movements over the different weeks. It is also noteworthy that the percentage of risky movements was the lowest on the farm using the 3-week system. This may be explained by the fact that the 3-week system allows for a better organization of the work throughout the weeks. In a 4-week system, there is one week with two main activities which demand extra work, i.e. insemination and farrowing, which could have led to a peak in the number of daily movements in that specific week of the BFS. Also, in the weaning week there were many movements, likely because sows had to be moved from the farrowing to the gestation/insemination unit and piglets from the farrowing to the nursery unit.

On farms A, B, and E, there were no significant differences in the percentage of risky movements according to the week of the BFS, meaning that the farmers applied a consistent working routine irrespective of the specific week of the BFS. Although there were no significant differences, the percentage of risky movements was high in all farms. Median values ranged from 9 to 38 %, indicating that farmers often do not adhere to the biosecurity standards. This implies that there is much room for improvement. On farms

C and D, there were significantly more risky movements in the weaning week. This was expected, as around the time of weaning there may be a lot of risky movements from the nursery to the farrowing unit.

In general, there were more movements on a weekday vs. a day on the weekend, and there are three possible explanations for this. The first one is that the BFS are well organized and most activities are planned on weekdays. In both the 3- and 4-week system, weaning takes place on a Thursday, sows are inseminated on Monday, Tuesday, and Wednesday, and sows farrow on Thursday, Friday, and Saturday. Second, in farms D and E, there were several workers and they might have not been all present on the farm during the weekend, resulting in less movements on the weekend. Third, the work on the farm could be done more efficiently during the weekend to save time for other non-farm-related activities. The percentage of risky movements on a weekday was higher on farms B, D, and E, but on farm C, this percentage was lower and more risky movements were made on a day during the weekend. On farm C, it is possible that the working routine was different during the weekend, and some extra violations to the correct working lines were made.

The total number of daily movements towards the farrowing and gestation/insemination unit was higher in the insemination and farrowing week, followed by the weaning week. The farrowing and gestation/insemination unit are places where much work is needed and farm staff possibly needed to visit these units more than once per day, e.g. for supervision at farrowing, treatment of suckling piglets, estrus detection, and insemination. For the nursery and fattening unit, the number of movements was similar regardless of the week of the BFS. In the absence of specific problems, these units were probably just visited for feeding and routine check of the animals.

The percentage of risky movements towards the farrowing unit and in some farms towards the gestation/insemination unit was higher in the weaning week, but we found no significant differences in the percentage of risky movements towards the nursery and fattening unit. Furthermore, the percentage of risky movements towards the nursery and fattening unit was in general much lower compared to the percentage of risky movements towards the farrowing and the gestation/insemination unit. The nursery and fattening unit were visited less frequently and these visits were probably better organized during the working day, facilitating the implementation of biosecurity principles and as such reducing the risk of making a risky movement.

The number of movements standardized by farm size towards the farrowing and gestation/insemination unit significantly differed according to the week of the BFS, while this was not the case for the number of movements standardized by farm size towards the nursery and fattening unit. In Table 6, showing the number of movements standardized by farm size, values below one can be noticed. The values in the table were obtained because standardization was done to allow a comparison between the farms. In some units, multiple detection points were present in one room. Furthermore, the movements were standardized per 100 sows for movements towards the farrowing and gestation/insemination unit, and all farms had more than 100 sows, leading to these values below one.

To our knowledge, the internal movement monitoring system Biorisk® is the first technology to verify movements of farm staff. There are only a few studies available where this technology has been used. Geurts et al. (2018) studied the association between the number of risky movements and the prevalence of porcine reproductive and respiratory syndrome virus in a farm; and Black et al. (2021) studied the association between movements and the number of weaned pigs per sow. In human medicine, similar technologies are already being used e.g. to monitor hand hygiene compliance in hospitals (Baslyman et al., 2015). The internal movement monitoring system allows real-time detection of farm staff. All information is immediately processed on the online platform. However, there are also some limitations to the system. The detection points should be plugged into a socket at all times and since the location of the detection points is crucial, in some farms extra sockets needed to be installed or extension cables were used. The range of the detection points is eight meters and goes through walls, so the time filter was needed to ensure that accidental detections were not registered. Furthermore, the system stands or falls by the dedication of the farm staff, as they should wear the beacon at all times. Regardless of these practical limitations, the internal movement monitoring system provided us with new and valuable information on the movements of farm staff in pig farms. The findings also complement the results of previous observational studies on biosecurity in pig farms.

Finally, besides the practical aspect, some ethical considerations are made. A previous study already raised some questions on data ownership, privacy, and cybersecurity concerning PLF (Wiseman et al., 2019). The Biorisk® system aims to understand movements of farm staff in order to improve biosecurity, not to check individual farm workers or accusing them of outbreaks. In case of unauthorized use, the system could violate privacy of farm staff and might cause difficulties for larger farms to find external staff willing to work on the farm. Furthermore, these data should not be used by the government or quality assurance schemes to verify if animals were checked daily.

CONCLUSION

The present study showed that there were a lot of (risky) movements on pig farms and that these movements varied according to week of the BFS, day of the week, and unit. This study creates awareness on movements of farm staff in pig farms, which is a first step in optimizing the working lines. It can lead to customized training for every farm based on objective data that show farm staff behavior and relating it to later health status and performance, aiming to promote a working culture of improving biosecurity, health and performance data-driven. Future research should provide insight into why specific risky movements occur and how these can be avoided to achieve a better biosecurity and higher health status on farms.

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CHAPTER 5 | DETERMINING THE CHARACTERISTICS OF FARMS THAT RAISE PIGS WITHOUT ANTIBIOTICS

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ABSTRACT

Reduced and responsible antimicrobial use (AMU) leads to a lower risk of developing antimicrobial resistance. Raised Without Antibiotics (RWA) is a certification label that is recognized in only a few countries, but it is often unclear what the specific criteria and characteristics of RWA farms are. The objectives of this study were to describe the criteria for a Belgian RWA program; to coach farms towards reduced AMU; to assess if it was possible to obtain and maintain the RWA status; and to determine differences between RWA and conventional pig farms. Pig farms (*n* = 28) were visited three times for the following reasons: (1) data collection, (2) farm-specific coaching (2 months later), and (3) evaluation (7 months later). AMU was followed from before the start of the study up to one year after the last visit. AMU, biosecurity (Biocheck.UGent), and farm characteristics of (non-)RWA farms were compared. RWA was defined as no antibiotics from birth until slaughter. Pigs requiring individual treatment received a special ear tag and were excluded from the program. The status of the farms varied over time, and the distribution of RWA vs. non-RWA was 10 – 18, 13 – 15, and 12 – 16, before intervention, after coaching, and after one year, respectively. For the non-RWA farms, there was a reduction in AMU of 61 %, 38 %, and 23 %, for the suckling piglets, fattening pigs, and sows, respectively, indicating that they were moving towards the RWA status. There were no significant differences in biosecurity status between RWA and non-RWA farms, but biosecurity improved in all farms throughout the study. RWA farms were smaller (median 200 sows) compared to non-RWA farms (median 350 sows). The 4-week system was used more in non-RWA farms, while the 3- and 5-week systems were used most often in RWA farms. This study showed that farmers could achieve and maintain the RWA status through farm-specific coaching related to prudent AMU and improved biosecurity.

INTRODUCTION

Antibiotics are used to tackle infectious diseases caused by bacteria; they can be used for therapeutic purposes and disease control in a herd (McEwen & Fedorka-Cray, 2002). However, there is a clear association between antimicrobial use (AMU) and antimicrobial resistance (AMR) (Chantziaras et al., 2014; Dorado-García et al., 2015; Holmer et al., 2019; Wall et al., 2016). At a national level, the use of specific antibiotics correlates to the level of resistance towards these antibiotics in commensal *Escherichia coli* isolates in pigs, poultry, and cattle (Chantziaras et al., 2014). The problem of AMR has been gaining attention over the years, since AMR can lead to therapeutic failure in both humans and animals, leading to increased morbidity and mortality. Currently, over 700 000 people worldwide die every year due to AMR. If no action is taken, it is estimated that by 2050 this number will increase to 10 million human deaths per year (Cosgrove, 2006; De Kraker et al., 2011; O'Neill, 2016). The problem of AMR should be addressed with a One Health approach including humans, animals, and the environment (European Commission, 2017). The reduction of antibiotic-resistant bacterial isolates in pig production can be obtained through coaching, better herd management, improved biosecurity, and prudent AMU or the restriction of AMU (Dewulf, 2018; Dorado-García et al., 2015; Postma, 2016; Postma et al., 2015).

Raised Without Antibiotics (RWA) in pig production is a concept that is recognized in only a few countries, such as Denmark, Poland, and the United States. In RWA production, pigs are raised without the use of any antibiotics from birth until slaughter (Baekbo, 2017; Cybulski et al., 2021; DANMAP, 2018; Lynegaard et al., 2021; Singer et al., 2019). In the United States, RWA is an independent certification that covers all animal source foods including meat, poultry, seafood, fish, dairy, and eggs. It is certified by the National Sanitation Foundation (NSF) (National Sanitation Foundation, 2023). In The Netherlands, there is also an antibiotic-free concept called 'Antibioticavrij Leven Garantie' (Duurzaam Varkensvlees, 2023). According to the Danish Integrated Antimicrobial Resistance Monitoring and Research Program (DANMAP), 51 pig farms in Denmark raised pigs without antibiotics in 2018 (DANMAP, 2018). Two studies, each investigating two RWA sow farms, further examined RWA production in Danish pig farms (Baekbo, 2017; Lynegaard et al., 2021). However, on a larger scale, it is unclear what the characteristics of RWA farms are, as well as which differences exist in comparison to conventional pig farms. Furthermore, the specific inclusion criteria for RWA production are not well specified in literature and the implementation of RWA in a larger number of farms with varying management and housing conditions requires further investigation.

Raising pigs without the use of antibiotics is challenging in terms of animal health and welfare. Especially after weaning, piglets are particularly susceptible to various infections, such as *E. coli*, causing post-weaning diarrhea or edema disease, or *Streptococcus suis*, causing bacterial meningitis, septicemia, and polyserositis. Previous studies have shown that most antibiotics in pig herds are used in nursery pigs (Baekbo, 2017; BelVet-SAC, 2022; Callens et al., 2012; Fairbrother & Nadeau, 2019). The decision as to whether or not to treat these bacterial infections with antibiotics is made after an evaluation performed by the herd veterinarian and the farmer and depends on the severity of the disease. In RWA production, the decision to treat can lead to exclusion from the program. However, RWA should not compromise animal welfare and the focus should be on the prevention of animal diseases.

The aims of this study were (1) to describe the criteria for a Belgian RWA program, (2) to guide pig farmers in the RWA program and assess if it was possible to achieve and maintain the RWA status, and (3) to determine the characteristics of the farms that succeeded.

MATERIALS AND METHODS

STUDY DESIGN

Belgian Pork Group is a network of abattoirs and companies active in cutting, deboning, and processing pig meat, and they commissioned the study to develop a product line of pigs raised without antibiotics. The project was presented at their annual meeting with pig farmers and farmers could apply voluntarily to participate in the project. To encourage participation in the project, collaborating RWA and non-RWA farms received a monthly incentive of € 250 and € 125, respectively. In total, 28 pig farmers applied to participate in the study. The geographical distribution of the farms is shown in Figure 1. All farms were located in Flanders, the northern Dutch-speaking part of Belgium.

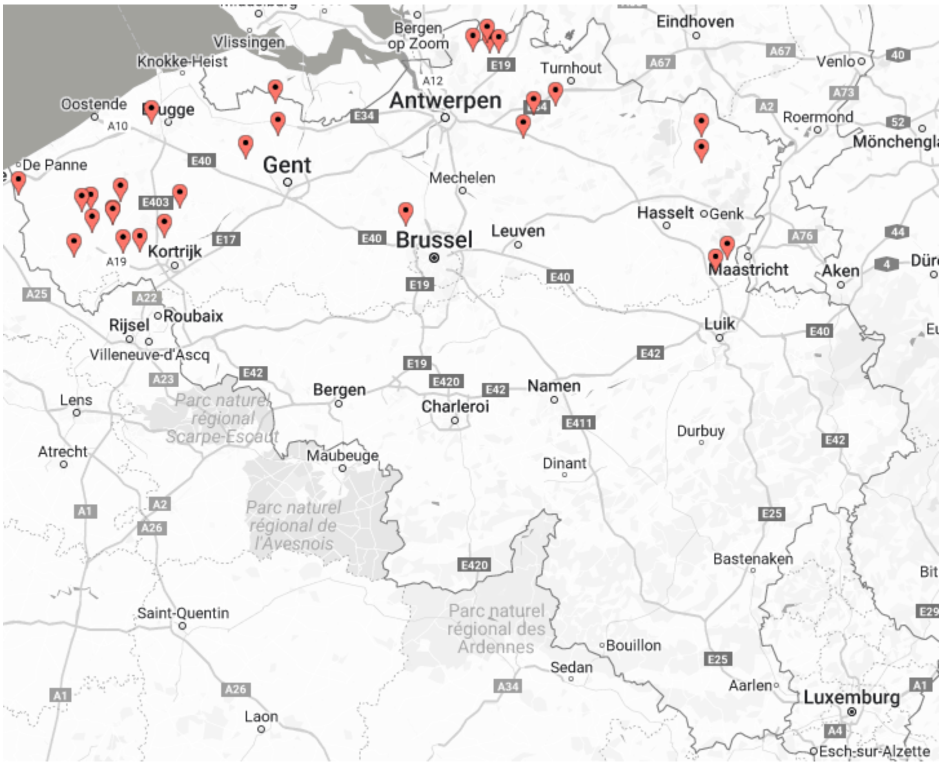

Figure 1. Map of Belgium with the geographical distribution of the farms.

Researchers of Ghent University performed the study. Due to practical reasons, the study was performed in two consecutive groups. The first group of farms (n = 16) was followed between 1 February 2018 and 30 September 2019; the second group (n = 12) was followed between 26 August 2019 and 30 June 2021. Both groups were approached and guided using the same methodology. All farms were visited three times by the same investigator. During the first visit, there was a herd inspection of the different animal categories and the overall farm infrastructure, combined with data collection (see next paragraph). Approximately two months later, a second visit was performed. The herd veterinarian was invited to discuss the situation on the farm together with the investigator and the farmer. It was determined as to whether or not the farms could start immediately in the RWA program or if a further reduction of AMU was initially required. Farm-specific recommendations were provided to support the farms in the RWA program or to guide farms towards RWA. During a third visit, approximately seven months after the second farm visit, the situation at the farm was evaluated again and compared to the situation of the first visit. AMU was monitored until one year after the third visit.

BIOSECURITY, FARM CHARACTERISTICS, AND PERFORMANCE

The biosecurity status of the farms was determined using the risk-based biosecurity quantification tool Biocheck.UGent (Biocheck.UGent, 2023). This tool allows for making an objective quantification of the biosecurity status of the farm. It is based on a questionnaire of 109 questions in 12 categories and results in a score between 0 and 100 %. Zero means a lack of any biosecurity measures, while 100 means perfect biosecurity. For external biosecurity, the following six categories were assessed: (1) purchase of breeding pigs, piglets, and semen; (2) transport of animals, removal of carcasses and manure; (3) feed, water, and equipment supply; (4) visitors and farmworkers; (5) vermin and bird control; and (6) location of the farm. For internal biosecurity, the following six categories were assessed: (1) disease management; (2) farrowing and suckling period; (3) nursery unit; (4) fattening unit; (5) measures between compartments, working lines, and use of equipment; and (6) cleaning and disinfection (Laanen et al., 2010; Laanen et al., 2013; Postma et al., 2016b).

Other farm characteristics - not included in Biocheck.UGent - were collected during the herd inspection (first farm visit) in a standardized way: name and contact details of the herd veterinarian, herd management characteristics including type of farm, batch farrowing system (BFS), sow breed, management and housing of the different animals, i.e. farrowing, gestation/insemination, nursery, fattening, and quarantine unit. A specific section was also reserved for all veterinary and non-veterinary treatments, including vaccination protocols, anti-parasitic treatment scheme (product, moment of treatment, and duration), and all feed and water additives.

Performance data for the past year were obtained from the herd management program during the first and third farm visit. The following information was collected for the sows and suckling piglets: number of weaned piglets per sow per year, farrowing index, weaning-to-estrus interval, pregnancy rate, replacement rate, liveborn piglets, pre-weaning mortality, and weaned piglets per litter. Reproductive data of the sows was available for most of the farms on the first visit, but fewer farms had follow-up information on the third visit. Therefore, the comparison of the performance of the first visit compared to the third visit was not made. For the nursery and fattening pigs, only 11 farms had information on mortality, average daily growth, and feed conversion ratio for the first visit, and only three farms could provide follow-up information for the third visit. Reasons for the loss of follow-up were the use of a different software program or a lack of time to reliably record or extract the data from the program. Therefore, this information was not further analyzed.

QUANTIFICATION OF ANTIMICROBIAL USE

Information on the AMU of the farms was provided by a nationally used database called AB Register. AB Register is an independent non-profit organization that deals with the registration of AMU for pigs, poultry, and dairy cows. All Belgian veterinarians have to register all antimicrobial use/supply for every farm in this database (AB Register, 2023). Based on the antibiotics used, the monthly BD100 was calculated. The BD100 is a standardized way to quantify the AMU and is nationally used in Belgium (AMCRA, 2022; Sarrazin et al., 2019; Timmerman et al., 2006). BD100 is the number of treatment days with antibiotics in 100 days or the percentage of treatment days with antibiotics and it is calculated for the suckling piglets, nursery pigs, fattening pigs, and sows. The numerator of the formula consists of the amount of antibiotics administered (expressed in milligram) and the long-acting factor (LA_{bel}), which corrects for products with an active duration longer than 24 hours. In the denominator, the Belgian defined daily dose animal was used ($DDDA_{bel}$). These values are defined based on the information in the summary of product characteristics (SPC) for each antibiotic. An overview of the specific values ($DDDA_{bel}$ and LA_{bel}) for each product was provided by the Belgian knowledge center on Antimicrobial Consumption and Resistance in Animals (AMCRA) (AMCRA, 2022). The total weight of animals at risk for treatment was the average number of animals at the farm multiplied by a standardized weight at treatment (see next paragraph). The number of days animals were at risk to receive treatment was also included. Regardless of the number of days at risk, the AMU was always converted to 100 days.

$$BD100 = \frac{\text{amount of antibiotics administered (mg)}}{DDDa_{bel} * \text{kg animal 'at risk'} * \text{number of days 'at risk'}} * LA_{bel} * 100$$

The average number of animals within each animal category was determined for each farm. For the suckling piglets, this was determined by multiplying the number of sows in a herd by 30, i.e. the average number of piglets per sow per year, divided by 12, i.e. months per year. The standardized weight of the pigs at treatment was defined as 4 kg, 12 kg, 50 kg, and 220 kg for the different animal categories, respectively. The number of days at risk was 30.42, i.e. the average length of a month.

AMU was compared to benchmarking values, which were determined for the different animal categories by the Belgian monitoring system. The benchmarking system includes alert and action values of the BD100 for the different animal categories. These values define three user categories. When the BD100 of an animal category is below the alert value, farms are considered to be low-user farms. They are in the safe zone and there is no need for action plans to reduce AMU. When the BD100 is between the alert and action value, the farms are considered to be intermediate users for an animal category. On these farms, extra attention should be paid to AMU and they should strive for a lower AMU. Finally, when the BD100 exceeds the action value, farms are considered to be high-user farms for an animal category. These farms should immediately take action to reduce AMU. In 2018, these benchmarking values were fixed by AMCRA (Table 1). In the future, these benchmarking values will be further tightened (AMCRA, 2019).

Table 1. Benchmarking values of the BD100 for the different animal categories (adapted from AMCRA, 2022). To raise pigs according to RWA criteria, the BD100 had to be below the alert value for at least three out of four animal categories.

	Alert value	**Action value**
Suckling piglets	2.00	11.00
Nursery pigs	14.00	51.00
Fattening pigs	2.70	9.00
Sows	0.28	1.65

AMU was determined for three periods per farm (Figure 2). The first period was approximately 14 months before the first farm visit (period A), the second period was the period between the first and the third farm visit (period B), and the third period was the period one year after the last farm visit (period C).

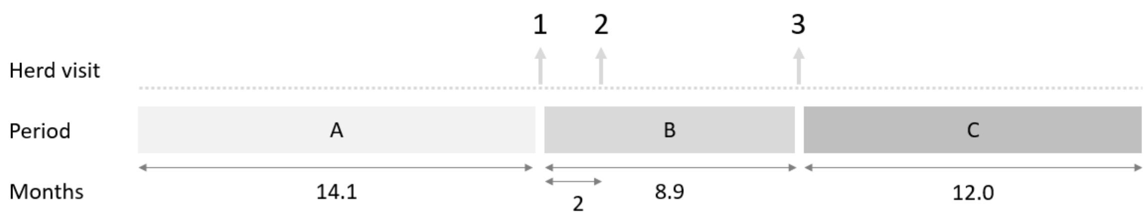

Figure 2. Timeline of the study. Three farm visits were performed, and antimicrobial use was determined for three different periods (A, B, and C).

CRITERIA OF THE BELGIAN RAISED WITHOUT ANTIBIOTICS PROGRAM

Before the start of the study, the inclusion criteria for RWA farms were defined to assure that RWA farms had a low AMU in all animal categories, did not apply any group treatments, and did not use antibiotics prophylactically. In those farms that were identified as RWA, an external company had to perform an annual audit to verify if the farms complied with all RWA criteria.

For the entire farm in an RWA program, prophylactic medication with antibiotics was not allowed. This was defined as the administration of a product to an individual animal or a group of animals without clinical signs to prevent the possible occurrence of an infection. Additionally, group treatments were not allowed. These were defined as any treatment from a therapeutic, metaphylactic, or prophylactic point of view, in which antibiotics were administered to a group of animals orally or parenterally. To comply with the RWA criteria, the BD100 had to be below the alert value of the Belgian benchmarking system for at least three out of four animal categories.

Within a farm that met the above criteria, only the animals that did not receive any antibiotics from birth until slaughter were labelled as RWA. Pigs originating from sows that received antibiotic treatment could still be raised according to the RWA criteria. If an animal required antibiotic treatment, individual treatments - i.e. parenteral, local, or oral treatments - were allowed. However, correct identification of the treated animals had to be performed carefully through the use of colored ear tags and forms indicating the antibiotic treatments and the identification and location of the treated animals. When pigs were moved to the next production stage, the treated animals had to be identified and housed separately. For example, suckling piglets that were treated with antibiotics in the farrowing unit had to be housed together in a separate pen in the nursery unit, and nursery pigs that received antibiotic treatment in the nursery unit had to be housed together in a separate pen in the fattening unit. Animals that had received antibiotic treatment were excluded from the RWA program and were slaughtered as conventional pigs. Figure 3 shows a flow chart with the criteria for farms to comply with the Belgian Raised Without Antibiotics program.

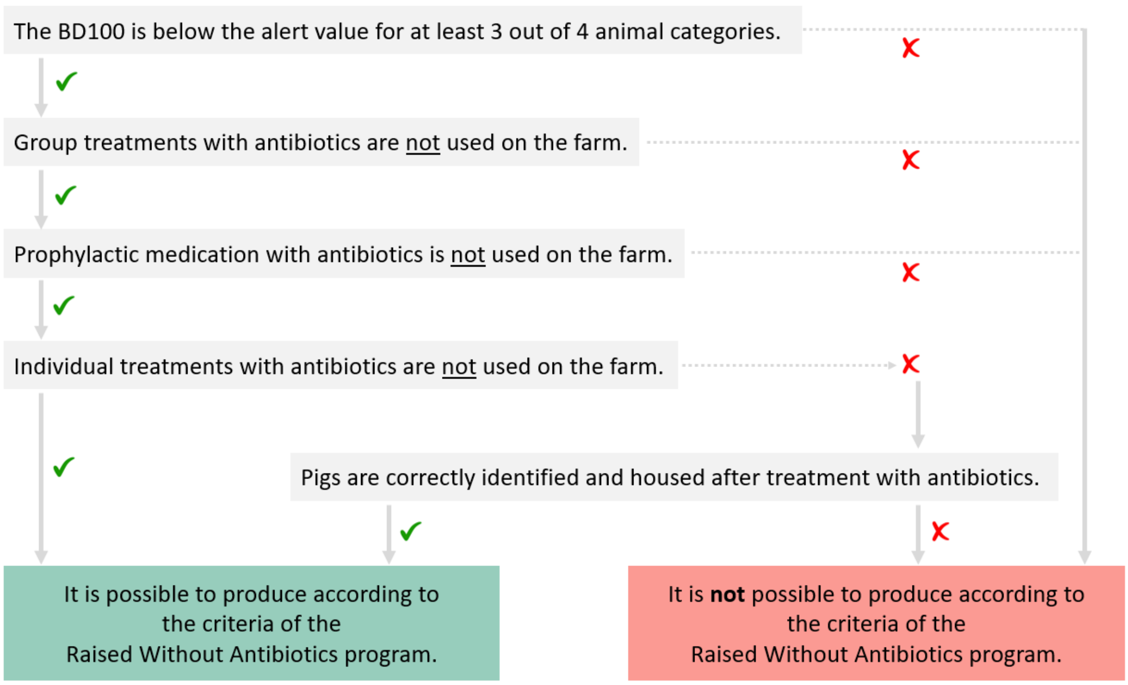

Figure 3. Flow chart to decide if a farm can produce according to the criteria of the Belgian Raised Without Antibiotics program.

DATA ANALYSIS

Descriptive statistics were performed for both the continuous and the categorical variables of the farm characteristics. Normality distribution was analyzed graphically via histograms and Q-Q plots. The mean ± SD was calculated for the normally distributed continuous variables, i.e. the weaning age of the piglets, number of pathogens against which animals were vaccinated, weaned piglets per sow per year, farrowing index, weaning-to-estrus interval, pregnancy rate, replacement rate, number of liveborn piglets, pre-weaning mortality, and the number of weaned piglets per litter. The median, minimum, and maximum values were determined for the not normally distributed continuous variables, i.e. number of sows, AMU (BD100), and biosecurity scores. Percentages were calculated for the categorical variables, i.e. type of farm, origin of the breeding gilts, BFS, possible castration of the boars, and sow breed.

A parametric independent samples t-test was used to analyze potential differences between groups for the normally distributed data. A non-parametric Mann-Whitney U test was used to analyze potential differences between groups for the not normally distributed data. The Levene's test was used for analyzing the equality of variances. A Fisher's exact test was used to assess differences between categorical variables. A nonparametric independent-samples Kruskal-Wallis test was used to analyze potential differences within groups for their AMU of the different periods. A multivariate analysis of variance (MANOVA) was performed to analyze potential effects of herd type (RWA vs. non-RWA), study group, and herd size on AMU for the different animal categories. A log transformation of the BD100 was performed to achieve normality distribution. In the model, BD100 was the dependent variable, herd type and study group were the fixed factors, and herd size was the covariate. A non-parametric Wilcoxon matched-pair signed-rank test was used to analyze potential differences within groups for their biosecurity status on the first farm visit compared to the third farm visit. *P*-values below 0.05 were considered to be statistically significant. All statistical analyses were performed using IBM® SPSS® Statistics for Windows Version 28 (IBM Corp., Armonk, NY, USA).

RESULTS

COMPLIANCE WITH THE BELGIAN RAISED WITHOUT ANTIBIOTICS PROGRAM

The mean (± SD) duration of periods A and B was 14.1 (± 1.0) and 8.9 (± 1.0) months, respectively. The duration of period C was the same for all farms, namely 12.0 months (Figure 2).

Taking into account the RWA criteria, farms could be categorized into two groups, i.e. RWA and non-RWA pig producers. Of course, this status could change over time and non-RWA farms could work towards RWA production. Therefore, the categorization was performed for the three periods, i.e. A, B, and C (Figure 4).

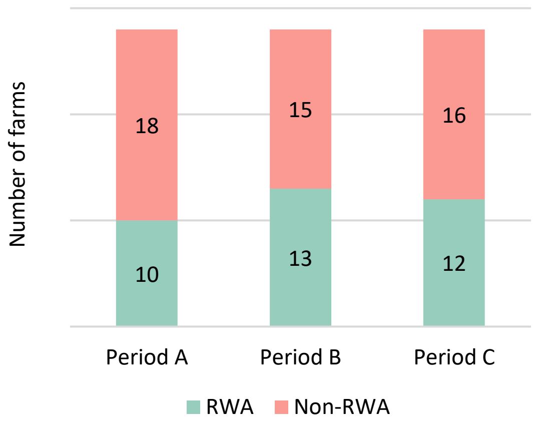

Figure 4. Number of farms producing according to the Raised Without Antibiotics (RWA) program and number of non-RWA farms in the different periods of the study.

Eight farms remained classified as RWA and 14 farms remained non-RWA during the entire study period. On six farms, the status varied over time (Table 2). These farms will be discussed in more detail in the following paragraphs.

Table 2. The status of the farms, i.e. (non-)RWA, could vary over time. For most farms, the status remained the same during the entire study. However, on six farms, the status varied in the different periods of the study (A, B, and C).

	Period A	**Period B**	**Period C**
Farm 1	non-RWA	RWA	RWA
Farm 2	non-RWA	RWA	RWA
Farm 3	non-RWA	RWA	RWA
Farm 4	non-RWA	RWA	RWA
Farm 5	RWA	RWA	non-RWA
Farm 6	RWA	non-RWA	non-RWA

Farm 1 was non-RWA in period A, but obtained and maintained the RWA status in periods B and C. Initially, the nursery pigs received amoxicillin trihydrate prophylactically (Rhemox premix, 15 mg per kg body weight (BW)), because there were problems with *S. suis* in the nursery unit. The prophylactic use of antibiotics is a reason for exclusion from the RWA program. Before, piglets from different sows were housed together in one pen in the nursery unit. After farm-specific coaching, the farmer changed the weaning practices and piglets of the same litter were housed together in a pen in the nursery unit. This allowed the farmer to stop with the prophylactic antibiotic medication of the nursery pigs and subsequently, the farm complied with the RWA criteria.

Farm 2 was non-RWA in period A. During this period, suckling piglets were treated with colistin sulphate (Colivet SF 500, 50,000 IU per kg BW) against neonatal diarrhea. Because of this treatment, the AMU of the suckling piglets exceeded the alert value. The alert value was also exceeded for the sows, because of antibiotic treatment in the farrowing unit with procaine benzylpenicillin (Peni-Kel 300,000 IE/mL, 21,000 IU per kg BW). Before, no separate clothing or boots were used for the different stables. Since pathogens can be transmitted indirectly via farm staff, it was advised to use different clothing and boots for the different animal categories. Especially the suckling piglets in the farrowing unit should be protected and the use of boots could help to avoid the transmission of pathogens. For periods B and C, there were no major health issues in the herd. Suckling piglets did not require treatment with colistin sulphate any longer, AMU did not exceed the alert value for this animal category, and the farm could produce according to RWA criteria.

Farm 3 was non-RWA in period A. During this period, the nursery pigs received group treatments with amoxicillin trihydrate (Octacillin 800 mg/g, 16 mg per kg BW) due to problems with *S. suis*. Even though AMU was below the alert value for all animal categories, the group treatment prevented them from producing according to the RWA criteria. In periods B and C, the farmer exchanged group treatments for individual treatments with amoxicillin trihydrate (Duphamox LA, 15 mg per kg BW) only for piglets with arthritis or meningitis. AMU remained below the alert value for all animal categories, making it possible for the farm to produce according to RWA criteria.

Farm 4 was non-RWA in period A because the AMU of two animal categories, i.e. fattening pigs and sows, exceeded the alert value. However, during period A, the farm applied complete depopulation and repopulated with specific-pathogen-free (SPF) sows. The sows were free of porcine reproductive and respiratory syndrome virus (PRRSV), swine influenza virus (SIV), *Mycoplasma hyopneumoniae*, *Actinobacillus pleuropneumoniae*, and *Glaeserella parasuis*. The result of this intervention was not immediately seen in the nursery and fattening pigs, since it took some time for the piglets of the SPF sows to reach the nursery and fattening unit. However, after some months, the health status of the farm significantly improved, resulting in fewer infections and less antimicrobial use. Therefore, the farm could produce according to RWA criteria in periods B and C.

Farm 5 was RWA in periods A and B. During these periods, only AMU of the suckling piglets exceeded the alert value and no group treatments were given. However, in period C, AMU of two animal categories, i.e. suckling piglets and sows, exceeded the alert value, resulting in the non-RWA status for period C. The main contributor to AMU in suckling piglets in period C was amoxicillin trihydrate (Duphamox LA, 15 mg per kg BW), which was administered against *S. suis* infections.

Farm 6 was RWA in period A, because there were almost no antibiotics treatments. However, at the end of period B, there were problems in the nursery unit with PRRSV and porcine circovirus type 2 (PCV2). The herd veterinarian decided to treat the nursery pigs with doxycycline hyclate (Doxyral 10% premix, 10 mg per kg BW) and amoxicillin trihydrate (Rhemox premix, 15 mg per kg BW) against secondary bacterial infections, leading to an increase of the BD100, exceeding the alert value. After these problems occurred in the nursery unit, all sows were vaccinated against PCV2 and a few weeks later all sows were intradermally vaccinated against PRRSV. In period C, the results of these vaccinations were not yet visible and the farm was still unable to produce according to RWA criteria.

CHARACTERISTICS OF RWA AND NON-RWA FARMS

To compare the characteristics of the RWA farms with the non-RWA farms, the status of the farms for period B, i.e. the period between visits 1 and 3, was considered. The comparison of the RWA (n = 13) and the non-RWA (n = 15) farms was made for farm characteristics, antimicrobial use, biosecurity, and performance.

FARM CHARACTERISTICS

The median (min. – max.) number of sows was 200 (85 – 300) for the RWA farms and 350 (180 – 1250) for the non-RWA pig farms ($p < 0.001$). The mean (± SD) weaning age of piglets was 24.9 (± 2.6) days for the RWA farms and 23.9 (± 2.9) days for the non-RWA pig farms ($p = 0.360$). Two of the RWA farms were SPF for different pathogens, e.g. *M. hyopneumoniae*, PRRSV, or SIV. Table 3 shows the other farm characteristics of the RWA and non-RWA farms separately.

RWA farms were more often single site farrow-to-finish farms compared to non-RWA farms ($p = 0.055$). RWA farms more often reared their own gilts compared to non-RWA farms ($p = 0.254$). There was a borderline significant association between the use of a specific BFS and farm status, i.e. (non-)RWA (p = 0.058). None of the RWA farms used the 4-week BFS, while this BFS was used most often on non-RWA farms. Possible castration of the boars did not seem to influence RWA status ($p = 0.320$). There was a borderline significant association between sow breed and RWA status ($p = 0.053$) and RWA farms seemed to use their own crossbred sows more often.

Table 3. Farm characteristics of the Raised Without Antibiotics (RWA) (n = 13) and the non-RWA pig farms (n = 15).

	RWA (*n* = 13)		**Non-RWA** (*n* = 15)	
	n	%	*n*	%
Type of farm				
Single site farrow-to-finish	11	85	7	47
Multiple sites	2	15	8	53
Origin of the breeding gilts				
Own rearing of breeding gilts	10	77	8	53
Purchasing of breeding gilts	3	23	7	47
Batch farrowing system				
1-week system	2	15	2	13
2-week system	1	8	0	0
3-week system	5	38	5	33
4-week system	0	0	6	40
5-week system	5	38	2	13
Castration of the boars				
Intact boars	2	15	6	40
Castration – chemical	4	31	5	33
Castration – surgical	7	54	4	27
Sow breed				
Belgian Landrace	1	8	2	13
Hypor	2	15	1	7
TN70	3	23	5	33
Danbred	0	0	3	20
Rattlerow Seghers	0	0	1	7
Danbred + Rattlerow Seghers	0	0	1	7
Danbred + Hypor	0	0	1	7
Own crossbred sows	7	54	1	7

RWA farms applied fewer vaccinations than non-RWA farms (Table 4). On the RWA farms, 46 % (6/13), 85 % (11/13), and 92 % (12/13) of the farms vaccinated the piglets, gilts, and sows, respectively. The farms not practicing gilt or sow vaccination were RWA for the entire study period. On the non-RWA farms, 80 % (12/15), 100 % (15/15), and 100 % (15/15) of the farms vaccinated the piglets, gilts, and sows, respectively.

Table 4. The number of pathogens (mean ± SD) against which piglets, gilts, and sows were vaccinated on the Raised Without Antibiotics (RWA) (*n* = 13) and the non-RWA pig farms (*n* = 15). *P*-values are provided for a comparison between RWA and non-RWA farms based on an independent samples t-test.

	RWA (*n* = 13)	**Non-RWA** (*n* = 15)	
	mean ± SD	mean ± SD	*p*-value
Vaccination piglets	0.7 ± 0.9	1.6 ± 1.1	0.025*
Vaccination gilts	3.5 ± 1.9	7.7 ± 2.1	< 0.001*
Vaccination sows	4.0 ± 1.8	6.3 ± 1.8	0.002*

* *P*-values below 0.05 were considered statistically significant

ANTIMICROBIAL USE

Table 5 shows the median (min. – max.) BD100 of the farms for the different animal categories for periods A, B, and C, for the RWA and the non-RWA farms separately.

First, a comparison of AMU of the different periods was made per animal category, for the RWA and non-RWA farms separately. No statistically significant differences were found. However, for the non-RWA farms, there was a reduction of the BD100 of period B compared to period A for the suckling piglets, the fattening pigs, and the sows, with a decrease of 61 %, 38 %, and 23 %, respectively.

Table 5. The median (min. – max.) BD100 of the farms for the different animal categories for the Raised Without Antibiotics (RWA) and the non-RWA pig farms. AMU was determined for three periods (A: 14 months before the first farm visit; B: between first and third farm visit; C: one year after third farm visit). The distribution of RWA vs. non-RWA farms was 10 – 18, 13 – 15, and 12 – 16, for periods A, B, and C, respectively. To raise pigs according to RWA criteria, the BD100 had to be below the alert value for at least three out of four animal categories.

	RWA						
	Alert value	Period A		Period B		Period C	
Suckling piglets	2.00	0.15	(0.00 – 26.10)	0.02	(0.00 – 10.10)	0.00	(0.00 – 1.28)
Nursery pigs	14.00	0.82	(0.35 – 15.56)	0.82	(0.00 – 29.18)	1.15	(0.05 – 10.27)
Fattening pigs	2.70	0.07	(0.00 – 0.95)	0.07	(0.00 – 1.86)	0.10	(0.00 – 0.56)
Sows	0.28	0.11	(0.00 – 0.83)	0.18	(0.00 – 2.23)	0.16	(0.00 – 1.02)
	Non-RWA						
	Alert value	Period A		Period B		Period C	
Suckling piglets	2.00	4.04	(0.00 – 78.40)	1.56	(0.00 – 34.48)	3.93	(0.00 – 24.73)
Nursery pigs	14.00	12.04	(0.02 – 75.90)	14.55	(1.20 – 97.78)	13.40	(0.00 – 88.78)
Fattening pigs	2.70	2.50	(0.11 – 9.66)	1.54	(0.00 – 5.48)	2.32	(0.00 – 11.20)
Sows	0.28	0.61	(0.00 – 5.98)	0.47	(0.02 – 12.71)	0.59	(0.00 – 5.34)

Second, we performed a MANOVA to evaluate the effect of herd type (RWA vs. non-RWA) on the AMU and corrected for the potential effects of study group and herd size. In none of the analyses, study group had a significant effect on AMU. The effect of herd size was in most cases not significant. Only in fattening pigs in period A ($p = 0.005$) and period B ($p < 0.001$), AMU was significantly higher on larger farms, whereas the AMU of sows in period B appeared to be significantly higher on smaller farms ($p = 0.037$) (Table 6). Regarding the herd type, i.e. RWA vs. non-RWA, there was a significant effect of RWA status on AMU of the following animal categories for the following periods: suckling piglets in period C ($p = 0.025$), nursery pigs in period B ($p = 0.009$) and period C ($p = 0.015$), fattening pigs in period A ($p < 0.001$) and period C ($p = 0.012$), and sows in period B ($p = 0.004$) and period C ($p = 0.032$).

Table 6. The median (min. – max.) BD100 of the animal categories where a significant effect of herd size on AMU was found. Farms were categorized into two groups; namely, farms with a herd size smaller than the median for the corresponding period and farms with a herd size equal to or larger than the median.

	Herd size < median	Herd size ≥ median
Fattening pigs (period A)	0.77 (0.00 – 9.66)	1.97 (0.01 – 8.92)
Fattening pigs (period B)	0.09 (0.00 – 4.24)	1.59 (0.00 – 5.48)
Sows (period B)	0.39 (0.00 – 12.71)	0.18 (0.00 – 4.21)

BIOSECURITY

Table 7 shows the median biosecurity scores (%) of the farms for the different categories of the Biocheck.UGent survey for the first compared to the third farm visit. The scores for the RWA and the non-RWA pig farms are shown separately. The overall external, internal, and total biosecurity scores did not significantly differ between the RWA and the non-RWA pig farms. However, within the two groups, i.e. RWA and non-RWA farms, the overall external, internal, and total biosecurity was significantly better on the third visit compared to the first visit. For both RWA and non-RWA farms, the overall internal biosecurity scores increased by 10 %. The biggest improvement was seen in the disease management of the RWA farms, with an increase in the score of 40 %. The biosecurity score of the fattening unit and measures between compartments, working lines, and use of equipment improved as well for both RWA and non-RWA farms.

Table 7. The median (min. – max.) biosecurity scores (%) of the farms for the different categories of the Biocheck.UGent survey for the Raised Without Antibiotics (RWA) (*n* = 13) and the non-RWA pig farms (*n* = 15). The survey was filled in during the first and third farm visit, jointly by the researcher and the farmer. *P*-values are provided for a comparison of biosecurity scores from the first and third farm visit based on a non-parametric Wilcoxon matched-pair signed-rank test.

	RWA (*n* = 13)					**Non-RWA** (*n* = 15)				
	Visit 1		Visit 3		*p*-value	Visit 1		Visit 3		*p*-value
External biosecurity	**66**	**(52 – 89)**	**71**	**(59 – 89)**	**0.005***	**70**	**(54 – 84)**	**72**	**(57 – 87)**	**0.002***
Purchase of breeding gilts, piglets, and semen	88	(56 – 100)	88	(60 – 100)	0.655	88	(76 – 100)	88	(78 – 100)	0.157
Transport of animals, removal of carcasses and manure	78	(39 – 87)	83	(70 – 90)	0.005*	78	(39 – 87)	83	(43 – 95)	0.012*
Feed, water, and equipment supply	33	(17 – 90)	37	(17 – 90)	0.180	37	(27 – 67)	40	(27 – 67)	0.180
Visitors and farmworkers	65	(35 – 100)	65	(47 – 100)	0.066	76	(65 – 100)	76	(65 – 100)	0.034*
Vermin and bird control	60	(30 – 100)	60	(30 – 100)	1.000	70	(30 – 100)	70	(30 – 100)	0.317
Location of the farm	70	(30 – 100)	70	(30 – 100)	1.000	40	(20 – 100)	40	(20 – 100)	1.000
Internal biosecurity	**48**	**(24 – 87)**	**58**	**(30 – 87)**	**0.005***	**53**	**(32 – 76)**	**63**	**(32 – 85)**	**0.018***
Disease management	40	(40 – 100)	80	(40 – 100)	0.025*	40	(40 – 100)	40	(40 – 100)	0.109
Farrowing and suckling period	64	(21 – 100)	71	(21 – 100)	0.068	57	(29 – 100)	71	(29 – 100)	0.109
Nursery unit	71	(36 – 100)	71	(43 – 100)	0.109	57	(14 – 86)	64	(14 – 86)	0.180
Fattening unit	64	(21 – 100)	79	(36 – 100)	0.042*	75	(36 – 100)	86	(36 – 100)	0.180
Measures between compartments, working lines, and use of equipment	32	(7 – 100)	50	(7 – 100)	0.018*	39	(18 – 86)	50	(18 – 86)	0.043*
Cleaning and disinfection	50	(0 – 98)	50	(0 – 98)	0.180	65	(20 – 95)	65	(20 – 95)	0.059
Total biosecurity	**56**	**(47 – 88)**	**64**	**(51 – 88)**	**0.005***	**62**	**(43 – 78)**	**65**	**(45 – 86)**	**0.002***

* *P*-values below 0.05 were considered statistically significant

PERFORMANCE

Table 8 shows the performance parameters of the RWA and the non-RWA farms. This information was collected on the first farm visit. No statistically significant differences were found. Nevertheless, on RWA farms there were less weaned piglets per sow per year compared to non-RWA farms.

Table 8. The mean ± SD values of the performance parameters on the Raised Without Antibiotics (RWA) (*n* = 13) and the non-RWA pig farms (*n* = 15). This information was collected on the first farm visit and not all parameters were available on all farms. *P*-values are provided for a comparison between RWA and non-RWA farms based on a parametric independent samples t-test.

	RWA (*n* = 13)		**Non-RWA** (*n* = 15)		
	n	mean ± SD	*n*	mean ± SD	*p*-value
Weaned piglets per sow per year	12	27.40 ± 3.60	15	28.97 ± 4.42	0.331
Farrowing index	12	2.34 ± 0.14	15	2.32 ± 0.12	0.669
Weaning-to-estrus interval (days)	8	6.21 ± 1.53	11	5.67 ± 0.73	0.375
Pregnancy rate (%)	9	94.24 ± 4.25	10	90.16 ± 5.38	0.086
Replacement rate (%)	10	44.10 ± 12.70	11	46.53 ± 10.79	0.641
Liveborn piglets	13	13.48 ± 1.18	15	14.41 ± 1.98	0.154
Pre-weaning mortality (%)	12	13.73 ± 5.28	15	13.29 ± 4.82	0.824
Weaned piglets per litter	12	11.62 ± 1.00	15	12.45 ± 1.51	0.115

* *P*-values below 0.05 were considered statistically significant

DISCUSSION

This study investigated the implementation of an RWA program applicable to Belgian pig farms. Pig farmers were guided in the program and the study showed that it was possible to achieve and maintain the RWA status. Furthermore, differences in farm characteristics between RWA and non-RWA herds were elucidated. All three aims of this study were met.

Pig farmers were able to apply voluntarily to this study. Therefore, the described farms are likely not representative of the whole population, as they can be assumed to be more interested and motivated to raise pigs with few antibiotics. On the other hand, interest and motivation are crucial to raise pigs according to the RWA criteria. Therefore, the presented results can be considered valid for the part of the population that qualifies for inclusion in this type of pig production.

As there is not yet a global agreement on the criteria of RWA production, there are differences in applied criteria between countries. The criteria for Belgian RWA production were drafted at the beginning of 2018. We defined that in RWA production, prophylactic use of antibiotics and group treatments for any reason, including prophylactic, were not allowed. In December of 2018, the new EU regulation on Veterinary

Medicinal Products was communicated and one of the goals of this new regulation was to strengthen the EU response to fight AMR. It was determined that prophylactic use of antibiotics should only be used in exceptional cases for administration to individual animals when the risk for infection is very high or the consequences are likely to be severe. Furthermore, the veterinarian should be able to justify the prescription of antibiotics, especially in the case of metaphylactic and prophylactic use. This new regulation bans the prophylactic use of antibiotics in groups of animals (European Council Regulation, 2018). Our criteria were in line with this legislation, which took effect in January of 2022.

In Denmark, the pig producer decides which piglets are suitable for RWA and provides them with a special ear tag before 4 days of age. If ear-tagged pigs receive a treatment with antibiotics, the special ear tag is then removed and the pig loses its RWA status (Baekbo, 2017; Lynegaard et al., 2021). In Poland, it is the other way around and pigs excluded from the RWA program receive an extra ear tag (Cybulski et al., 2021). In the Belgian RWA program, all piglets born in a farm that fulfills the RWA criteria start automatically as RWA. If a treatment with antibiotics is required, treated pigs get a special ear tag and lose their RWA status. The location of the antibiotic treated pigs must be known at all times, by using a form indicating identification and location of antibiotic-treated animals. This was included, because there is always the risk that a treated animal loses its ear tag. If this happens, it is then made clear by using the form which animals should be excluded from the RWA program.

According to DANMAP, 51 pig farms in Denmark raised RWA pigs in 2018 (DANMAP, 2018). In 2018, there were in total 1613 farms with sows in Denmark, meaning that 3 % of the Danish sow farms were producing RWA pigs in 2018 (Statistics Denmark, 2022). In Belgium, only the farms of this study are known to produce according to RWA criteria. In 2020, there were 1649 sow farms in Belgium. This would mean that currently only 0.7 % of the Belgian sow farms produce according to RWA criteria (StatBel, 2020).

Antibiotic-free strategies, i.e. the complete restriction of all antibiotics, might be beneficial in reducing AMR, but antibiotic treatments are sometimes really necessary to treat animals that are clinically diseased due to infections with bacterial pathogens. Not treating such animals would have a negative impact on animal performance, farm profitability, and, last but not least, animal welfare (Karavolias et al., 2018). In a study of Baekbo (2017), discontinuation of routine antibiotic treatment because of the initiation of an RWA program resulted in an increased incidence of umbilical hernia, diarrhea, and arthritis, and the piglets had a slightly lower weight at the end of the nursery unit, illustrating that the introduction of an RWA program is not always feasible or warranted (Baekbo, 2017). On the other hand, in the same study, the RWA program did not seem to have a negative impact on the overall productivity of the sow farms and RWA fattening pigs showed fewer lesions at slaughter, i.e. chronic pneumonia, hernia, and abscesses, compared to non-RWA pigs (Baekbo, 2017).

In a survey in the United States, 88 % of the respondents with RWA experience and 98 % of the respondents with no RWA experience believed that RWA production slightly or significantly worsens animal health, animal welfare, and food safety, even though antibiotic treatment of sick animals was allowed. Most respondents agreed that more stringent health and welfare auditing is needed when animals are raised without antibiotics, to ensure a good follow-up of animal health (Singer et al., 2019). In the RWA program described in the current study, it is possible to treat sick animals at all times, but consequently, such treated pigs lose their RWA status. If the antibiotic treatment does not cause a high increase of the BD100 and the alert value is not exceeded, the farm maintains its RWA status. As such, the animal health and welfare should not be negatively influenced by the program.

In general, most of the antibiotic treatments are administered in nursery pigs (DANMAP, 2018; SDa, 2020). The study of Baekbo (2017) on RWA production in two sow farms showed that the nursery period was challenging and only 69 to 75 % of the pigs that were initially selected to participate in the RWA program were still RWA after the nursery period. The most frequently occurring problems were diarrhea, arthritis, and umbilical hernia. Only 38 to 58 % of the ear-tagged pigs were finally slaughtered as RWA (Baekbo, 2017). In the study of Lynegaard et al. (2021) on RWA production on two farms, similar percentages were found and 64 and 68 % of the pigs reached the end of the nursery period without any antibiotic treatment (Lynegaard et al., 2021). This is in line with the findings of the present study, where most antibiotic treatments were administered to the nursery pigs. The exact percentage of pigs that received antibiotic treatment in our study was not known. However, this was expected to be limited, as farms had to comply with the criteria of the RWA program and AMU in general had to be low.

There was an important reduction in AMU for the suckling piglets, fattening pigs, and sows in the non-RWA farms from period A to period B, likely a result of the fact that these farms were guided towards RWA. In the RWA farms, no significant reductions of AMU were observed between period A and B, likely because AMU was already very low, hence there was not much room for further reduction.

The effect of study group, herd size, and herd type on AMU was evaluated. We found that AMU of the fattening pigs increased with a larger herd size, which is in agreement with several previous studies (Backhans et al., 2016; Van der Fels-Klerx et al., 2011). However, for the sows, we found that larger herds appeared to use less antimicrobials. Yet, the difference in absolute values of AMU in the sows was quite limited. Therefore, it is questionable as to whether the observed effect is also biologically relevant.

Given the importance of the proverb “Prevention is better than cure”, the focus should be on the prevention of infectious diseases by improving hygiene and preventing the spread of infection by biosecurity measures (Dewulf et al., 2018; O'Neill, 2016). In our study, significant improvements in the biosecurity score were established and the biggest improvements were made for the internal biosecurity. For both RWA and non-RWA farms, the overall internal biosecurity scores increased by 10 %. The biggest improvement was seen in the disease management of the RWA farms, with an increased score of 40 %. Also, in the study of Baekbo (2017), biosecurity measures were mentioned: enhanced hygiene measurements in the stables and when handling the pigs, the all-in/all-out principle, and changing footwear internally in the farm when different compartments were visited (Baekbo, 2017).

RWA farms were smaller (median 200 sows) compared to non-RWA farms (median 350 sows). On farms with fewer sows, the groups of nursery and fattening pigs will be smaller. Since direct contact with live animals is the main disease transmission route and a high stocking density can cause a strong rise in the infection pressure (Dewulf et al., 2018), it can be expected that the infection pressure decreases on farms with smaller groups of pigs. Subsequently, there is less of a need to treat animals with antibiotics. Therefore, RWA could be mainly feasible on small farms.

An increased weaning age of piglets reduces the risk of developing post-weaning disease, e.g. *E. coli* diarrhea (Fairbrother & Nadeau, 2019), and has been associated with a lower AMU (Caekebeke et al., 2020; Postma et al., 2016a). In our study, the weaning age was on average slightly higher on RWA farms (24.9 ± 2.6 days) compared to non-RWA farms (23.9 ± 2.9 days). This is also in line with the Danish approach, where increasing weaning age was part of the RWA strategy (Baekbo, 2017). Remarkably, there were no RWA farms using a 4-week BFS. A possible explanation could be that farms working in a 4-week system wean their piglets usually one week earlier (at max. 21 days) than farms working, for example, in a 3-week system (weaning at 28 days of age). A higher weaning age leads to more robust, resilient, and heavier piglets that have adapted better to solid feed, resulting in a smoother transition at weaning. Furthermore, the performance of piglets weaned at four weeks of age is better (Barceló, 2009). This could lead to less of a need for treatment with antibiotics.

The distribution of farms that did own rearing of breeding gilts and farms that purchased them was 77 – 23 % and 53 – 47 % for RWA and non-RWA farms, respectively. Even though this difference was not significant, it is nonetheless interesting that RWA farms seemed to purchase fewer breeding gilts. Generally, it is assumed that purchasing breeding gilts is a biosecurity risk, as through the introduction of new animals new pathogens also may be introduced, which may lead to more diseases and subsequently, a higher need for antibiotic treatment (Dewulf et al., 2018).

There were also no significant differences between RWA and non-RWA farms with regard to the used sow breed. Nevertheless, it is remarkable that none of the RWA farms used the highly prolific Danbred sows, which can give birth to an average of 17.6 liveborn piglets per litter (Schild et al., 2020). Along with these large litters, the average birth weight of the piglets is lower compared to sows that produce fewer piglets. Piglets with a lower birth weight show a higher risk of stillbirth and pre-weaning mortality (Quiniou et al., 2002). Moreover, piglets with a lower birth weight show reduced post-weaning performance (Quiniou et al., 2002). These piglets might be more vulnerable to diseases in the nursery or fattening unit, thus having a higher chance of being treated with antibiotics later in life.

In our study, there was no statistically significant difference between RWA and non-RWA farms for possible castration of the boars, i.e. intact boars, chemical, or surgical castration. However, there were some non-RWA farms using antibiotics prophylactically with surgically castrated boars. These farms could not be included in the RWA program, since RWA farms were not allowed to use antibiotics prophylactically.

We found that RWA farms applied fewer vaccinations compared to non-RWA farms. A smaller proportion of RWA farms vaccinated their piglets, gilts, and sows, and they vaccinated against significantly fewer pathogens. This might seem contradictory since a reduction in AMU can be obtained by increased vaccination (Dupont et al., 2017; Postma et al., 2015; Postma et al., 2017). A possible explanation might be that the health status on RWA farms was higher, resulting in a lower need for vaccination against different pathogens. Furthermore, there were two SPF farms amongst the Belgian RWA farms. On these farms, specific pathogens were absent against which vaccination often occurs in conventional pig farms, e.g. *M. hyopneumoniae*, PRRSV, or SIV. This finding supports an earlier study that indicated that farms vaccinating against more pathogens have a higher AMU from birth until slaughter (Postma et al., 2016a).

Finally, there were no statistically significant differences in the performance parameters of RWA and non-RWA farms, suggesting that raising pigs without antibiotics does not necessarily compromise the performance of the pigs. However, we only have information on performance parameters of the sows and pre-weaning piglet mortality, as other information such as mortality rate, average daily growth, and feed conversion ratio of the nursery and fattening pigs was lacking. However, a previous study by Postma et al. (2017) showed that it was possible to reduce AMU without jeopardizing the production parameters (Postma et al., 2017).

CONCLUSION

This study showed that it was possible to achieve and maintain the RWA status through farm-specific coaching related to prudent AMU and biosecurity. Characteristics of farms that succeeded were determined, and differences between RWA and non-RWA farms were elucidated. The criteria of the RWA program are clearly described in this study and can be used in other studies. The characteristics of RWA farms can be used to estimate whether farms are suitable to raise their pigs without antibiotics. Further research is needed to reveal the feasibility of RWA on a larger scale.

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CHAPTER 6 | GENERAL DISCUSSION

A systematic reduction of antimicrobial use (AMU) will result in a gradual decrease in antimicrobial resistance (AMR). It will also prevent the selection of new resistance types and will slow down the spread of current resistance genes (Dewulf, 2018). Currently, pigs can still be medicated therapeutically or metaphylactically with antimicrobials. The new regulation on Veterinary Medicinal Products has strengthened the European Union (EU) legislation in an attempt to fight AMR. To this end, prophylactic use of antimicrobials can only be used in exceptional cases and is banned for medicating groups of animals (European Council Regulation, 2018). Also other efforts can be made to reduce or even completely eliminate AMU. In this framework, the Raised Without Antibiotics (RWA) concept is interesting. This general discussion will critically reflect on biosecurity, more in particular the introduction procedures of breeding gilts (Chapter 3) and movements of farm staff (Chapter 4), the reduction of AMU, and the feasibility of RWA (Chapter 5). These topics will be discussed in two sections, namely: (1) the challenges that might be faced when reducing (or eliminating) AMU, and (2) the way forward with a focus on some crucial factors, including biosecurity, to reduce AMU.

1. THE CHALLENGES OF RAISING PIGS WITHOUT ANTIMICROBIALS

There are some considerations to be made when raising pigs without antibiotics. First, you must ensure that the pigs remain healthy and productive, which is a challenge, since there are studies indicating that this might be difficult. Therefore, reducing or eliminating AMU must always be accompanied with other measures, such as increased biosecurity. Second, in case of RWA, you have to make sure that the product will get sold. The consumers must be well informed regarding RWA and the possible consequences in terms of animal welfare, food quality and safety, and production costs. These challenges will be further discussed in the next paragraphs.

1.1 MAINTAINING HEALTH AND PRODUCTION

When reducing AMU, it is important to address unnecessary use first. Prophylactic use of antimicrobials is still routinely used on some farms (Sarrazin et al., 2019; Sjölund et al., 2016), possibly because farmers use this as a kind of insurance policy to minimize specific risks. Proper prevention could reduce the need for prophylactic treatments. Afterwards, the overall health of a farm can be improved and curative antimicrobial treatments can also be reduced. In the past, it has already been shown that herd-specific coaching related to prudent AMU and biosecurity decreased both prophylactic and curative AMU (Postma et al., 2017). All animals should be kept healthy through prevention, optimal management, good biosecurity, and proper housing and feeding. Animals that are sick should be treated, but this should involve accurate diagnostics to detect the etiologic agent to select the proper treatment and avoid new disease outbreaks (Dewulf, 2018).

A study was conducted to gain insight in the opinion of veterinarians and producers on RWA. Veterinarians felt that, in some cases, the RWA label had priority over animal health and welfare (Singer et al., 2019). In some studies, RWA negatively impacted pig health, welfare, and production (Baekbo, 2017; Dee et al., 2018), confirming the opinion of veterinarians. For poultry, different studies examined the impact of RWA on the birds and in general, there was a negative impact on gut health, bird performance, and animal welfare (Gaucher et al., 2015; Karavolias et al., 2018; Smith, 2011). However, Karavolias et al. (2018) recognized that other management practices, i.e. other factors than conventional vs. RWA production, were not further examined. According to the study of Karavolias et al. (2018), shifting to RWA production in poultry should include some production changes, such as reduced stocking density and increased downtime between production cycles in a barn (if not yet sufficient).

In some studies, the impact of RWA was not limited to animal health and welfare only. In broiler production, RWA resulted in a less efficient production, i.e. average daily gain (ADG), feed conversion ratio (FCR), and mortality, which in turn led to a decrease in edible meat. To compensate for these losses, more broilers were needed, impacting the environment and economic viability due to associated increases in feed and water requirements and increased manure production. Subsequently, this could also lead to increased prices for the consumer (Cervantes, 2015; Salois et al., 2016). Dee et al. (2018) investigated the impact of RWA on pigs that were experimentally challenged with porcine reproductive and respiratory syndrome virus (PRRSV). They found that RWA pigs were more clinically diseased, had a lower ADG, a higher FCR, and a higher mortality rate, which could in turn also lead to an increased number of pigs to meet the demand for animal protein. However, we must be careful when extrapolating findings from this study to practice, since RWA production is not recommended on farms with serious health problems.

In our study, we found no statistically significant differences in pre-weaning piglet mortality of RWA and conventional farms. Unfortunately, data on ADG, FCR, and mortality of nursery and fattening pigs was lacking (Chapter 5). Nonetheless, it was shown in a previous study that AMU could be reduced without jeopardizing performance parameters (Postma et al., 2017); and another study found that ADG and mortality were similar on conventional and RWA pig farms (De Bruyn, 2019). When reducing or eliminating AMU without further actions, there might be an impact on health, production, and subsequently the economic viability. However, RWA production should be accompanied with sufficient biosecurity measures and other alternatives such as vaccination. In that case, raising pigs without antibiotics might be feasible without negatively affecting health or production. Furthermore, the Belgian RWA program allows individual antimicrobial treatment for pigs that would require it, ensuring animal health.

1.2 GETTING THE PRODUCT SOLD

Consumers' perception is important, as they should ultimately buy the product. For the RWA concept, brand reputation and willingness to pay (WTP) are crucial. First, it's important to distinguish between citizens and consumers. Citizens are individuals who are member of a country or community. They may be concerned about animal welfare and can advocate for animal rights and supporting legislation; they can participate in public opinion formation. On the other hand, consumers are individuals who engage in purchasing, influencing the marketplace. They have the power to make choices that can influence the treatment of animals in livestock production. Over the past years, citizens' and consumers' concern regarding animal welfare in current livestock production systems has increased. Especially women, pet owners, younger, higher-educated, and higher-income people were more concerned. However, decent knowledge on livestock production and animal welfare is lacking. Consequently, consumers' view on animal welfare is mainly driven by (mis)perception rather than facts (Alonso et al., 2020).

Consumers can have different perceptions on several aspects of RWA production. The first one is animal welfare. Several studies have shown that consumers believe RWA significantly improves animal welfare (Bradford et al., 2022; Denver et al., 2021; Singer et al., 2019), or that welfare-friendly products originate from animals receiving less or no antibiotics (Clark et al., 2016). However, consumers are not aware that banning antimicrobials might have negative effects on animal health and welfare. When eliminating AMU, simultaneous changes in management must be established, otherwise there could be an increase in animal suffering (Goddard et al., 2017). The second aspect is food quality and safety. On this topic, conflicting results are found in literature. Sometimes consumers perceive RWA pork of lesser quality (Bradford et al., 2022), while other studies found that RWA was perceived as safer and higher-quality food (Singer et al., 2019). The third aspect is production costs. Raising pigs without antimicrobials may come at an additional cost, which is also the perception of consumers (Bradford et al., 2022; Denver et al., 2021). This additional cost might discourage consumers from purchasing RWA pork, leading us to the final aspect, namely WTP. Also regarding WTP, studies contradict each other. In some studies, WTP for RWA products was high (Denver et al., 2021; Patel et al., 2020), while in others it was rather low (Bradford et al., 2022). WTP depends on different factors, e.g. animal species. WTP for welfare-friendly products is from low to high: pigs, fish, broilers, laying hens, dairy cows, and beef cows; thus, especially for pigs, consumers have a lower WTP for welfare-friendly products (Alonso et al., 2020). However, for RWA, the impact of species on WTP was not yet investigated. WTP also depends on socio-demographic variables, e.g. education, gender, age, but also on the frequency of pork consumption (Bradford et al., 2022; Denver et al., 2021). In Belgium, farms received a monthly incentive to participate in the RWA program (Chapter 5). However, we did not look into the economic aspect of RWA production. Further research is needed to determine if RWA production includes additional costs and if a monthly incentive could be sufficient to cover these possible extra costs for the farmer.

2. THE WAY FORWARD: CRUCIAL FACTORS TO RAISE PIGS WITHOUT ANTIMICROBIALS

As previously mentioned, it is clear that you cannot simply stop using antimicrobials in conventional pig production systems without changing and improving the surrounding circumstances. As a consequence, there are some crucial factors to consider when you want to raise pigs without antimicrobials: (1) farmers' motivation, (2) the role of animal health professionals, (3) checking and improving biosecurity, and (4) monitoring, benchmarking, and reducing (or eliminating) AMU. These topics will be discussed more in detail in the next paragraphs.

2.1 FARMERS' MOTIVATION

Reducing AMU should be accompanied with other measures, such as increased biosecurity. However, in order to make successful and long-term changes, the first crucial factor is farmers' motivation. The ADKAR® model consists of five elements that are important for change to succeed (Chapter 1) (Houben et al., 2020). The model can be used for both biosecurity and antimicrobial stewardship. By applying the ADKAR® model, stakeholders in pig production can navigate through the stages of change, increasing the likelihood of successful implementation and compliance with biosecurity, and subsequently reduce their AMU (Caekebeke, 2021). Unfortunately, we did not apply the ADKAR® model in our study. This approach would ensure that all aspects of the change process, from raising awareness to building capacity, are addressed systematically, leading to a more successful reduction (or elimination) of AMU on pig farms. Moreover, it could help identify where things might go wrong. Different tools could be used to improve the elements of the ADKAR® model, e.g. performing surveys (Chapter 3) or (demonstration) projects (Chapters 4 and 5), communication about these projects, financial incentives, articles, training courses, e-learning modules, and testimonials.

2.2 THE ROLE OF ANIMAL HEALTH PROFESSIONALS

Reducing AMU requires a comprehensive approach. Besides motivation of the farmer, it is important to involve animal health professionals, e.g. to give advice or to perform studies to find and evaluate alternative strategies. Also herd veterinarians play a crucial role to support and provide knowledge, as they have a bond of trust with farmers. So a joint effort from all animal health professionals is needed to reduce AMU or to raise pigs without antimicrobials. These professionals include veterinary practitioners, scientists, nutritionists, technical consultants from pharmaceutical companies, and veterinary consultants or advisors. Animal health professionals can collaborate in different ways.

- Animal health professionals can collaborate to educate pig farmers and other stakeholders about AMR and the importance of prudent AMU. This education can also include information on alternative strategies, such as biosecurity.

- Collaboration of animal health professionals can lead to developing guidelines and best practices regarding prudent AMU. A good example of this is the establishment of AMCRA in Belgium (AMCRA, 2022).
- Animal health professionals can develop surveillance systems to monitor AMU. In Belgium this led to the development of AB Register and Sanitel-Med. But also in other countries there are already surveillance systems (AACTING consortium, 2019).
- Research initiatives aimed at finding alternative strategies for reducing AMU are very important. This can include biosecurity or other management practices to promote animal health and reduce diseases requiring antimicrobial treatment.
- Also legislation and policy are important to reduce AMU. Belgian legislation restricts the use of red antimicrobials. An example of Belgian policy is the covenant to fight AMR, where the Belgian Federal Government and various animal production partners were involved.

These are just a few examples of how animal health professionals can collaborate. By collaborating and working across disciplines, animal health professionals can make significant strides in reducing AMU in pig production. In our study, herd veterinarians were also closely involved in the coaching process (Chapter 5). Other studies found that veterinarians were perceived as the most reliable source of information on disease control (Alarcon et al., 2014; Nöremark et al., 2009). Therefore, it is important to involve them in this process. But also the role of other animal health professionals should be reviewed and enhanced, to further facilitate support, guidance, and establishment of effective herd biosecurity and health programs.

2.3 BIOSECURITY: CHECK AND IMPROVE

When motivated farmers are surrounded by animal health professionals, they can start focusing on disease prevention and biosecurity, in order to reduce AMU on farms. To do this consistently, the check – improve – reduce method can be used (Dewulf, 2018; Postma, 2016). To do so, it is important to keep the following recommendations in mind:

- Follow the correct order: First check the biosecurity, then improve it, and finally reduce AMU. Do not start reducing AMU immediately, without evaluating the farm's biosecurity and management first.
- Stick to the plan: Failing to plan is planning to fail. Try to make biosecurity easy and fun to sustain the efforts.
- One step at a time: Try to start with some 'quick wins' to keep up the motivation. Then you can continue with more difficult or drastic changes.
- Make full use of hard-to-ignore reminders: e.g. visual material such as posters, colors, or physical barriers. This can help farm staff and visitors to comply with the biosecurity measures.

- Assure to have the proper equipment: Sufficient and good equipment per compartment can make work more pleasant.
- Introduce some competition: Biocheck.UGent allows to compare yourself to the country average and to follow up your own progress, which could help to even further improve.
- Provide training programs: Education and practical workshops can help farmers and farm staff to put theory into practice. In this, repetition is key.

2.3.1 CHECK

When assessing biosecurity on a farm, it is important to distinguish between structural and operational biosecurity. Structural biosecurity deals with physical factors, e.g. farm layout, number of hygiene locks, presence of showers, air filtration systems, etc. The structural biosecurity can be assessed by surveys and audits. For example, by using Biocheck.UGent, poor practices can be identified and improved (Biocheck.UGent, 2023). The operational biosecurity depends on routine procedures to prevent introduction and spread of disease within the farm, e.g. taking a shower, changing clothing and footwear, washing hands, etc. (Vaillancourt, 2023). The operational biosecurity can also be evaluated using surveys such as Biocheck.UGent, but sometimes it can be helpful to go a step further and verify the adherence to biosecurity measures by using cameras or sensors (Vaillancourt, 2023).

Although biosecurity measures can be theoretically in place, there is often lack of compliance, due to three different main reasons (Smith et al., 2023; Vaillancourt, 2023):

- Environment: design of the farm, economic constraints, lack of time, difficulty to apply suggested measures, absence of necessary material.
- People: beliefs, attitudes, perceptions, education, experience, personality traits.
- Knowledge: lack of knowledge on biosecurity, inadequate training, inadequate communication (incoherence), absence of valid audits, lack of incentives.

So the effectiveness of biosecurity measures largely depends on the compliance of farmers, farm staff, and visitors. A study in poultry production used hidden cameras to identify biosecurity flaws on eight poultry farms. After 883 visits by 102 different individuals, 44 biosecurity flaws were observed. The most common flaws were related to respecting the areas (clean vs. contaminated), washing hands, and wearing coveralls and boots. These flaws suggest that there is a lack of biosecurity knowledge and proper training could improve knowledge and actually complying to biosecurity principles (Racicot et al., 2011). In a follow-up, visible cameras and audits were used to check the compliance to biosecurity measures on the farms. Visible cameras significantly improved changing boots and respecting areas for the short-term. However, after six months, the compliance decreased again. Bimonthly audits did not improve medium-term biosecurity compliance (Racicot et al., 2012b).

This thesis focused on the two main disease transmission routes, i.e. live animals (purchasing of breeding gilts, Chapter 3) and people (movements of farm staff, Chapter 4). Both studies gained insight in compliance to biosecurity measures, created awareness, and revealed biosecurity flaws, clearly showing which improvements can be made on these farms. In Chapter 3, the introduction procedures of breeding gilts were evaluated using Biocheck.UGent, with some additional questions. The results showed that the optimal introduction procedures of breeding gilts were only applied on 10 % of the farms. On the remaining farms, one or more biosecurity flaws related to the introduction procedures were present, which could potentially lead to pathogen introduction on the farm. In Chapter 4, the real-time monitoring system Biorisk® identified the movements of farm staff by using beacons and detection points (Animal Data Analytics, 2023). Monitoring movements of farm staff in pig farms is an important biosecurity aspect and can provide valuable insights. By monitoring the movements, you can ensure that proper working lines are followed, which helps minimizing the risk of disease introduction. Furthermore, it allows to analyze work patterns and identify areas for improvement in terms of workflow and time management. While monitoring movements of farm staff has those benefits, it is essential that privacy and autonomy of employees is respected. Clear communication, well-defined policies, and appropriate data protection measures should be in place to address any concerns and maintain a positive work environment.

2.3.2 IMPROVE

Once the biosecurity status and possible flaws on the farm are known, it is possible to improve the situation. On-farm biosecurity measures are well-described and have been shown to be very effective. However, biosecurity measures should be strictly applied, the resources must be available and transparent, and people's knowledge and motivation must be increased (Vaillancourt, 2023). Nöremark and Sternberg-Lewerin (2014) investigated whether certain biosecurity resources were available for veterinarians and other farm visitors. They reported that on some farms there was no adequate protective clothing or coverall and boots were dirty or wrong size. There were also some obstacles for washing hands, i.e. no availability of water or soap. In human medicine, compliance to hand hygiene in hospitals was improved by different interventions, such as intensive training, frequent and visible monitoring, immediate feedback, and providing real-time data to managers (Walker et al., 2014). So it is clear that education and training of farm staff is important. Furthermore, farm staff should also be motivated to apply biosecurity measures. Also other personality traits were associated with compliance to biosecurity and understanding personality patterns could help in the way farm staff is trained (Racicot et al., 2012a; Vaillancourt, 2023).

While technologies can be used to check biosecurity, they can also be effective for improving biosecurity by increasing the compliance, e.g. by real-time feedback and alarms notifying biosecurity breaches (Racicot et al., 2022). These systems could be further modified, e.g. by linking them to the door lock, so if biosecurity measures were not fulfilled in the hygiene lock, the stable door will not open (Vaillancourt, 2023).

As previously mentioned, introduction of breeding gilts and movements of farm staff include a high risk of pathogen transmission. Therefore, implementing biosecurity measures related to these two transmission routes will have a great impact on decreasing introduction and spread of pathogens on pig farms. Chapter 3 revealed that a lot of improvements could be made regarding the introduction procedures of breeding gilts. Own rearing of breeding gilts could be beneficial in terms of biosecurity. Moreover, Chapter 5 showed that most of the RWA farms reared their own breeding gilts. However, for own rearing of breeding gilts, other factors should also be considered, such as cost, farm size, expertise, and workload. Currently, Belgian legislation does not require a quarantine period of newly purchased animals if farmers only want to transport fattening pigs to the slaughterhouse; if they want to transport other pigs, e.g. nursery pigs or sows, a quarantine period of four weeks is required for the newly purchased animals. This legislation seems to focus on not spreading pathogens from the farm to the outside world and not vice versa, while the quarantine period is even more important for the farmer to protect the animals that are already present on the farm and to allow proper acclimation of the purchased animals. Improving the introduction procedures of breeding gilts involves a combination of factors, including legislation, communication, and training. Legislation should require a quarantine duration of purchased breeding gilts of minimum four weeks to protect the sow herd and to allow proper acclimation of the gilts. Furthermore, the optimal introduction procedures of breeding gilts should be communicated to farmers. The survey that we performed was a good way to raise awareness among participating farmers. After the study, an article on the topic was published in a magazine called 'Varkensbedrijf', which was an opportunity for all farmers to read the results of the study and to gain knowledge on the optimal introduction procedures. Chapter 4 revealed that the percentage of risky movements on farms ranged from 9 to 38 %. While farmers claimed to organize their working consistently starting with the young animals and then continuing the work in the older animals, this was not always the case and high percentages of risky movements were observed. There was a discrepancy between the answer on the Biocheck.UGent question regarding working lines and the actual situation. Farm structure and design could help optimizing the working lines. However, when it comes to older farms, it is not easy. For new farms, the stables should be built in a way that respecting the working lines is logical and easy.

2.4 ANTIMICROBIAL USE: MONITOR, BENCHMARK, REDUCE OR RAISE WITHOUT ANTIBIOTICS

Only if the previous three factors are fulfilled, i.e. motivated farmer, involvement of animal health professionals, and improved biosecurity, farmers can start reducing AMU.

2.4.1 MONITORING AND BENCHMARKING OF ANTIMICROBIAL USE

"To measure is to know"; therefore, monitoring and benchmarking AMU are crucial to establish antimicrobial stewardship programs and to measure their effectiveness. Legislation and other initiatives regarding monitoring of AMU in Europe and Belgium are thoroughly described in Chapter 1. In our study, we used the BD100 to calculate AMU for the different animal categories on the participating pig farms (Chapter 5). This way, AMU was calculated in a standardized way, allowing comparison between the different periods, which allowed us to determine if AMU was evolving in a favorable way.

Besides monitoring AMU, also benchmarking of AMU is important. It allows farmers and veterinarians to evaluate their AMU practices and it has some specific advantages:

- Benchmarking provides a basis for comparison. It allows farmers to compare their AMU to other farms. This can help to identify areas where AMU can be reduced. It might also encourage farmers to do better than other farms.
- Benchmarking facilitates monitoring trends over time. This way, efforts that were made to reduce AMU can be identified. Farmers might become motivated to see that their efforts have paid off.
- Benchmarking encourages transparency in AMU, demonstrating a commitment to responsible AMU.
- Benchmarking can help identify opportunities for improvement. It encourages farmers to adopt best practices for responsible AMU.

The Belgian benchmarking system includes alert and action values of the BD100 for the different animal categories, i.e. suckling piglets, nursery, fattening, and breeding pigs (Chapter 1). These values will be further adjusted and in AMCRA's 'Vision 2024' the reduction path is determined (Table 1) (AMCRA, 2019). These reduction paths help in reducing AMU. The color codes from the AMCRA user categories, i.e. green – orange – red, could be referred to as traffic light systems. This visual representation might encourage farmers to reduce AMU and to stay in the green zone. Furthermore, specific action should be taken when AMU exceeds the alert or action values. Farms with a long-term or repeated high AMU should be supported by an external coach, which has financial consequences (Chapter 1). In Chapter 5, the Belgian benchmarking system was also used to determine if farms could raise pigs according to the RWA program, since the first criterium of RWA was that the BD100 had to be below the alert value for at least three out of four animal categories.

Table 1. Thresholds of the BD100 reduction path 2021 – 2024 for the suckling piglets, nursery, fattening, and breeding pigs in Belgium (adapted from AMCRA, 2019).

	Alert value			**Action value**		
	Jan 2021	**Jan 2023**	**Jan 2024**	**Jan 2021**	**Jan 2023**	**Jan 2024**
Suckling piglets	2.00	2.00	2.00	10.00	6.00	5.00
Nursery pigs	14.00	14.00	14.00	50.00	40.00	30.00
Fattening pigs	2.70	2.70	2.70	9.00	6.00	6.00
Breeding pigs	0.28	0.28	0.28	1.65	1.65	1.65

Table 2 shows the distribution of the farms in the different user categories for the different animal categories at the end of 2022. For all animal categories, there were approximately between 60 and 70 % low-user and between 3 and 8 % high-user farms. The alert values will remain stable for the coming years, thus the percentage of low-user farms should stay quite stable. Since the alert values are used in the Belgian RWA program (Chapter 5), it is not expected that RWA farms would be affected by the reduction paths.

In contrast, the action values of the suckling piglets, nursery, and fattening pigs were adjusted in January 2023. With the current results, it would be expected that 5 to 15 % of the farms would become high-user farms, i.e. in the red zone, after this adjustment of the action values (BelVet-SAC, 2023). Although the median BD100 has decreased for all animal categories over the past years, the situation is worrying regarding the reduction paths. The action values of the suckling piglets and nursery pigs will even further decrease in January 2024. If no action is taken, the high-user farm percentage will further increase.

Table 2. Percentage of farms in the different user categories at the end of 2022 for the different animal categories (adapted from BelVet-SAC, 2023).

	Low-users (BD100 < alert)	**Alert-users** (alert < BD100 < action)	**High-users** (BD100 > action)
Suckling piglets	71,0 %	23,1 %	5,8 %
Nursery pigs	64,5 %	28,5 %	7,0 %
Fattening pigs	70,4 %	21,6 %	8,0 %
Breeding pigs	59,7 %	35,9 %	3,4 %

2.4.2 REDUCE OR RAISE PIGS WITHOUT ANTIBIOTICS

The ultimate goal is to improve pig's health and to eventually raise pigs with as few antimicrobials as possible or even without antimicrobials. The distinction between the two concepts is important. Reducing AMU is something everyone should strive for, while RWA might not be feasible on all farms. On some farms, it might be possible to raise pigs without antimicrobials immediately, while on other farms a reduction of AMU is necessary first.

Legislation pushes farmers towards a reduced AMU. The European legislation bans the prophylactic use of antimicrobials in groups of animals and has strict guidelines for the prophylactic use of antimicrobials in individual animals, making it almost impossible to use antimicrobials preventively of routinely (European Council Regulation, 2018). In addition, the AMCRA reduction path ensures that all farms have to reduce their AMU (AMCRA, 2019). However, for successful reduction of AMU, the intrinsic motivation of farmers should be increased.

The RWA concept is also known in different species, such as pigs, poultry, beef, and dairy cattle (Singer et al., 2019). Labeling meat products could help consumers to express their preferences (Centner, 2016). Nonetheless, it was found that RWA claims were sometimes false and animals were administered antimicrobials anyway (Price et al., 2022). Therefore, when farmers produce for a certain label, it is important to perform thorough audits to prevent false claims.

The motivation to raise animals without antimicrobials may vary. There is concern about AMR, but also other factors such as legislation, animal health and welfare, trading opportunities, and consumer preferences may play a role (Singer et al., 2019). Singer et al. (2019) surveyed veterinarians and producers in different animal species, i.e. broiler, turkey, swine, beef, and dairy cattle, about their experience and perception of RWA. The main factor contributing to the decision to raise animals without antibiotics was market-driven, namely to fulfill a client/customer request. In contrast, the main factor contributing to the decision to raise animals conventionally was the fact that the respondents were concerned about the negative impact of RWA on animal health and welfare.

When comparing AMR on conventional and RWA farms, there are conflicting results. Chekabab et al. (2021) found a reduced frequency of AMR genes on RWA pig farms compared to conventional farms, while other studies found similar AMR levels on conventional and RWA cattle (Schmidt et al., 2021; Vikram et al., 2017) and pig farms (Tunsagool et al., 2021). Another study found even more resistance genes in the nasal microbiome in sows that were housed on RWA farms compared to conventional farms (Alvarado et al., 2022). We should keep in mind that reducing AMU or RWA concepts are a tool to reduce AMR, but proving the association between AMU and AMR is sometimes difficult and often requires long-term studies. Nevertheless, previous studies have shown that a decrease in AMU resulted in significantly lower

AMR levels (Callens et al., 2018; Dorado-García et al., 2016; Tang et al., 2017). This also becomes clear when looking at the BelVet-SAC report, where a decrease in AMU is reflected in a decrease of AMR (BelVet-SAC, 2023).

For pigs, the RWA concept is only known in a few countries and the inclusion criteria or specific characteristics of RWA farms are not well specified (Chapter 1). Chapter 5 clearly describes inclusion criteria for the Belgian RWA program. These guidelines might serve as inspiration for other studies or countries. We showed that RWA was possible through farm-specific coaching related to prudent AMU and biosecurity. Chapter 5 also describes specific characteristics of the RWA farms in our study and the differences with (conventional) reducing farms. On RWA farms the herd size was on average smaller than 200 sows. Smaller farms might have a lower disease pressure or could find it relatively easier to manage and monitor individual pigs for health issues. RWA farms were mainly single site farrow-to-finish farms with own rearing of crossbred sows, which could be beneficial in terms of biosecurity, decreasing the opportunity of pathogen introduction into the farms. None of the RWA farms in our study were applying a 4-week batch farrowing system, indicating that piglet's lower weaning age might have negative effects on their health, resulting in a higher need for antimicrobial treatment after weaning. Also none of the RWA farms used high prolific Danbred sows, a sow breed producing large litters with a lower average birth weight. RWA farms applied fewer vaccinations than non-RWA farms, which could indicate a higher health status of the farms and less need to vaccinate animals. Unfortunately, we found no significant association in biosecurity status between RWA and non-RWA farms.

Taking into account the findings from our study and the characteristics of RWA farms, could RWA be feasible on all farms? Unfortunately, the study was too limited to determine whether RWA could be feasible on a larger scale. But in my opinion, there are already some type of farms where RWA does not seem possible, such as farms in a 4-week system with a low weaning age. Besides the fact that these farms are probably uncapable of producing pigs according to the RWA criteria, there are also some other considerations to be made, e.g. the fact that piglets on these farms are systematically weaned at a too young age (max. 21 days). Farms with high prolific sows might also not be able to produce according to the RWA criteria, as these sow breeds have large litters, low births weights, and reduced post-weaning performance.

3. FINAL CONCLUSIONS AND FUTURE PERSPECTIVES

Even though great strides have already been made to reduce AMU, the road towards complete RWA pig production in Europe remains long. When looking at Belgium, RWA production is already feasible in some farms. However, this is on a very small scale and a herd-specific approach is necessary. Motivated farmers with high awareness and desire could be coached to improve their knowledge and ability to raise pigs without antimicrobials by focusing on biosecurity and preventive veterinary medicine. The characteristics of RWA farms from Chapter 5 can be used to estimate whether farms could be suitable for raising pigs without antibiotics. However, further research is needed to check the feasibility of RWA on a larger scale. On farms with certain characteristics, e.g. farms working in a 4-week system or with high prolific sows, it seems highly unlikely for the RWA program to succeed.

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SUMMARY

Worldwide, pigs are one of the most commonly raised animals for meat production. To meet the rising demand for animal protein, livestock production has been intensified during the past decades. This intensification was accompanied by increased stocking density, leading to a more efficient pathogen transmission between live animals, resulting in increased antimicrobial use (AMU). Unfortunately, AMU results in selection pressure for antimicrobial resistance (AMR). In Europe and in Belgium, efforts are made to reduce AMU. Also legislation has changed and monitoring of AMU became obligatory, and prophylactic medication of groups of pigs was prohibited. Since 2011, AMU in all pig categories, i.e. suckling piglets, nursery pigs, fattening pigs, and breeding pigs, has decreased in Belgium. However, further efforts must be made.

The **general introduction** (**Chapter 1**) shows the evolution of intensive livestock production, the discovery of antimicrobials, and the reasons for the development of AMR. AMU in pig production is described in detail, including the important risk factors. Furthermore, the situation in Europe and Belgium and efforts to reduce AMU in pig production are elucidated, including the 'Raised Without Antibiotics' concept (RWA). Chapter 1 focuses on biosecurity as an alternative for AMU, since biosecurity improves animal health and production, reduces AMU, and subsequently improves the profitability of the farm. Other alternatives, such as vaccination, feed (additives), optimizing the stable climate and housing conditions, and coaching are also reviewed.

The overall **aim** of this thesis (**Chapter 2**) was to improve animal health, to create awareness on biosecurity, and to reduce antimicrobial use in pig farms. The specific objectives were to investigate the introduction procedures of purchased breeding gilts, assess the movements of farm staff, describe criteria for a Belgian RWA program, evaluate if farms could achieve and maintain this status, and finally identify possible differences between RWA and non-RWA farms.

Contact with live animals, including breeding gilts, is the most risky transmission route for pathogens. Therefore, some precautionary measures should be taken when purchasing breeding gilts. A study on the purchasing policy, quarantine and acclimation practices of breeding gilts in Belgian pig farms is shown in **Chapter 3**. Fifty-seven percent of the farms purchased breeding gilts and there was a lot of variation in the frequency of purchase and the age at which gilts were purchased. On 95 % of the purchasing farms, a quarantine unit was used. On most of the farms, the quarantine was located on the farm itself (internal quarantine). The median (min. – max.) duration of the quarantine period was 42 (14 – 140) days. Only 10 % of the farms complied with the optimal introduction procedures of breeding gilts, as described in literature. The results from Chapter 3 show that in many farms, practices related to purchasing, quarantine, and acclimation of breeding gilts could be improved to maintain optimal biosecurity.

In terms of internal biosecurity, it is important to separate different age groups in a pig farm and consistently use specific working lines when visiting the barns: (1) hygiene lock, (2) farrowing, (3) gestation/insemination, (4) nursery, (5) fattening, (6) quarantine unit, and (7) cadaver storage. The movements of farm staff, which is an important route of pathogen transmission, was investigated in five pig farms and described in **Chapter 4**. The total number of movements differed according to the week of the batch farrowing system (BFS) and was highest during the insemination and farrowing week. The percentage of risky movements was influenced by the week of the BFS for two farms and was highest around weaning. The percentage of risky movements varied significantly between farms, ranging from 9 to 38 %. There were more movements on a weekday compared to a weekend day. There were more movements towards the farrowing and gestation/insemination unit in the insemination and farrowing week compared to other weeks of the BFS, but the week of the BFS had no impact on movements towards the nursery and fattening unit. The results from Chapter 4 show that there were many (risky) movements on pig farms and that these movements varied according to week of the BFS, day of the week, and unit. This study created awareness on the importance of staff movements in pig farms, which could be a first step in optimizing working lines and decreasing risky movements.

The criteria of the Belgian RWA program are described in **Chapter 5**. RWA was defined as no antibiotic use from birth until slaughter. Pigs requiring individual treatment were identified with a special ear tag and excluded from the program. Twenty-eight pig farms were included in the study and coached towards reduced AMU. The status of the farms varied over time and the distribution of RWA vs. non-RWA was 10 – 18, 13 – 15, and 12 – 16, before intervention, immediately after coaching, and one year later, respectively. For the non-RWA farms, there was a reduction in AMU of 61 %, 38 %, and 23 %, for the suckling piglets, fattening pigs, and sows, respectively, indicating that they were moving toward the RWA status. There were no significant differences in biosecurity status between RWA and non-RWA farms, but biosecurity improved in all farms throughout the study. RWA farms were smaller (median 200 sows) compared to non-RWA farms (median 350 sows). The 4-week BFS was more applied in non-RWA farms, while the 3- and 5-week BFS were mainly used in RWA farms. The results of Chapter 5 show that farmers could achieve and maintain the RWA status through farm-specific coaching related to prudent AMU and improved biosecurity.

Finally, the **general discussion** (**Chapter 6**) focusses on the current situation in Europe and Belgium regarding AMU and the RWA concept. Of course, there are some challenges we are facing when it comes to reducing AMU or raising pigs without antibiotics: animal health and production, and the consumer. In order to raise pigs without antibiotics, the following measures are needed: (1) to coach farmers, (2), to work together with all animal health professionals, (3) to check and improve biosecurity, and (4) to monitor, benchmark, and reduce AMU. This is necessary for all stakeholders to improve animal health and to move towards raising pigs without antibiotics.

SAMENVATTING

Wereldwijd zijn varkens een van de meest gehouden dieren voor de vleesproductie. Om aan de stijgende vraag naar dierlijke eiwitten te voldoen, is de veeteelt de afgelopen decennia intensiever geworden. Deze intensivering ging gepaard met een hogere bezettingsdichtheid, wat leidde tot een efficiëntere overdracht van ziekteverwekkers tussen levende dieren, wat resulteerde in een verhoogd antibioticagebruik. Helaas leidt antibioticagebruik tot selectiedruk voor antimicrobiële resistentie. In Europa en België worden inspanningen geleverd om het antibioticagebruik te verminderen. Ook de wetgeving is veranderd en het monitoren van antibioticagebruik werd verplicht en profylactische groepsbehandeling met antibiotica werd verboden. Sinds 2011 is het antibioticagebruik in alle diercategorieën voor varkens, d.w.z. zuigende biggen, gespeende biggen, vleesvarkens en zeugen, afgenomen in België. Verdere inspanningen zijn echter nodig.

De **algemene inleiding** (**Hoofdstuk 1**) toont de evolutie van de intensieve veehouderij, de ontdekking van antimicrobiële stoffen en de redenen voor de ontwikkeling van antibioticaresistentie. Antibioticagebruik in de varkenshouderij wordt in detail beschreven, inclusief belangrijke risicofactoren. Verder worden de situatie in Europa en België en de inspanningen om antibioticagebruik in de varkensproductie te verminderen toegelicht, waaronder het concept 'Raised Without Antibiotics' (RWA). Hoofdstuk 1 richt zich op bioveiligheid als een alternatief voor antibioticagebruik, aangezien bioveiligheid de diergezondheid en productie verbetert, antibioticagebruik vermindert en ook de economische rendabiliteit van het bedrijf verbetert. Andere alternatieven, zoals vaccinatie, voeder(additieven), het optimaliseren van het stalklimaat en de huisvesting, en coaching worden ook besproken.

De **doelstelling** van dit proefschrift (**Hoofdstuk 2**) was het verbeteren van de diergezondheid, het creëren van bewustzijn over bioveiligheid en het verminderen van het antibioticagebruik op varkensbedrijven. De specifieke doelstellingen waren het onderzoeken van de maatregelen bij introductie van aangekochte fokgelten, het beoordelen van de bewegingen van medewerkers of varkensbedrijven, het beschrijven van criteria voor een Belgisch RWA-programma, evalueren of bedrijven deze status konden bereiken en behouden, en tot slot het vaststellen van mogelijke verschillen tussen RWA- en niet-RWA bedrijven.

Contact met levende dieren, waaronder fokgelten, houdt een hoog risico in om ziekteverwekkers over te dragen. Daarom moeten er enkele voorzorgsmaatregelen worden genomen bij de aankoop van fokgelten. Een studie over het aankoopbeleid, de quarantaine en de adaptatie van fokgelten in Belgische varkensbedrijven wordt weergegeven in **Hoofdstuk 3**. Zevenenvijftig procent van de bedrijven kocht fokgelten aan en er was veel variatie in de frequentie en leeftijd waarop gelten werden aangekocht. Op 95 % van de aankopende bedrijven werden fokgelten eerst in een quarantaine geplaatst. Op de meeste bedrijven bevond de quarantaine zich op het bedrijf zelf (interne quarantaine). De mediaan (min. – max.) duur van de quarantaineperiode was 42 (14 – 140) dagen. Slechts 10 % van de bedrijven voldeed aan de optimale introductiemaatregelen voor fokgelten, zoals beschreven in de literatuur. De resultaten van

Hoofdstuk 3 laten zien dat op veel bedrijven de maatregelen met betrekking tot aankoop, quarantaine en adaptatie van fokgelten verbeterd kunnen worden om zo naar een optimale bioveiligheid te streven.

Voor de interne bioveiligheid is het belangrijk om verschillende leeftijdsgroepen op een varkensbedrijf te scheiden en consequent specifieke looplijnen aan te houden bij een bezoek aan de stallen: (1) hygiënesluis, (2) kraamstal, (3) dracht/dekstal, (4) biggenbatterij, (5) vleesvarkensstal, (6) quarantaine en (7) kadaveropslag. De looplijnen van personeel, een belangrijke manier voor de overdracht van pathogenen, werd onderzocht op vijf varkensbedrijven en beschreven in **Hoofdstuk 4**. Het totale aantal bewegingen verschilde naargelang week van het wekensysteem en was het hoogst tijdens de dek- en werpweek. Het percentage risicovolle bewegingen werd voor twee bedrijven beïnvloed door de week van het wekensysteem en was het hoogst tijdens speenweek. Het percentage risicovolle bewegingen varieerde aanzienlijk tussen de bedrijven, nl. van 9 tot 38 %. Er waren meer bewegingen op een weekdag in vergelijking met een weekenddag. Er waren meer bewegingen naar de kraam- en dek/drachtstal in de dek- en werpweek in vergelijking met andere weken van het wekensysteem, maar week van het wekensysteem had geen invloed op de bewegingen naar de biggenbatterij en vleesvarkensstal. De resultaten van Hoofdstuk 4 laten zien dat er veel (risicovolle) bewegingen waren op varkensbedrijven en dat deze bewegingen varieerden naargelang week van het wekensysteem, dag van de week, en stal. Deze studie creëerde bewustzijn over het belang van bewegingen van personeel op varkensbedrijven, wat een eerste stap zou kunnen zijn in het optimaliseren van looplijnen en het verminderen van risicovolle bewegingen.

De criteria van het Belgische RWA-programma worden beschreven in **Hoofdstuk 5**. RWA werd gedefinieerd als geen gebruik van antibiotica vanaf geboorte tot slacht. Varkens die een individuele behandeling nodig hadden, werden geïdentificeerd met een speciaal oormerk en uitgesloten van het programma. Achtentwintig varkensbedrijven werden opgenomen in de studie en begeleid naar een verminderd antibioticagebruik. De status van de bedrijven kon veranderen in de loop van de studie en de verdeling van RWA vs. niet-RWA was respectievelijk 10 – 18, 13 – 15, en 12 – 16, voor de interventie, meteen na de coaching, en een jaar later. Voor de niet-RWA bedrijven was er een vermindering in antibioticagebruik van 61 %, 38 % en 23 % voor respectievelijk de zuigende biggen, vleesvarkens en zeugen, wat aangeeft dat ze evolueerden in de richting van de RWA-status. Er waren geen significante verschillen in bioveiligheid tussen RWA- en niet-RWA bedrijven, maar de bioveiligheid verbeterde op alle bedrijven gedurende de studie. De RWA bedrijven waren kleiner (mediaan 200 zeugen) dan de niet-RWA bedrijven (mediaan 350 zeugen). Het 4-wekensysteem werd meer toegepast op niet-RWA bedrijven, terwijl het 3- en 5-wekensysteem vooral werden gebruikt op RWA bedrijven. De resultaten van Hoofdstuk 5 laten zien dat veehouders de RWA-status konden bereiken en behouden door bedrijfsspecifieke begeleiding met betrekking tot voorzichtig antibioticagebruik en verbeterde bioveiligheid.

Tot slot focust de **algemene discussie** (**Hoofdstuk 6**) op de huidige situatie in Europa en België met betrekking tot antibioticagebruik en het RWA-concept. Natuurlijk zijn er een aantal uitdagingen waar we voor staan als het gaat om het verminderen van antibioticagebruik of het grootbrengen van varkens zonder antibiotica, zoals diergezondheid, het milieu en de consument. Om varkens groot te brengen zonder antibiotica zijn de volgende maatregelen nodig: (1) veehouders coachen, (2) samenwerken met verschillende deskundigen op vlak van diergezondheid, (3) bioveiligheid controleren en verbeteren en (4) antibioticagebruik monitoren, benchmarken en verminderen. Deze stappen zijn noodzakelijk voor alle belanghebbenden om de diergezondheid te verbeteren en te evolueren naar het grootbrengen van varkens zonder antibiotica.

CURRICULUM VITAE

Elise Bernaerdt was born in Eeklo (Belgium) on 7th of August in 1992. After successfully completing her secondary education (Sciences and Mathematics) at Sint-Laurens Zelzate in 2010, she started Veterinary Medicine at Ghent University (Belgium). In 2016, she obtained her Master's degree in Veterinary Medicine (main subject Pigs, Poultry & Rabbit).

In July 2016, she started as a pig practitioner at veterinary practices De Linde in Retie (Belgium) and De Grensstreek in Bladel (The Netherlands), where she was responsible for sampling, pregnancy diagnosis, and routine herd visits on pigs farms.

In February 2018, she started as a doctoral student in the unit of Veterinary Epidemiology (Department of Internal Medicine, Reproduction and Population Medicine) under supervision of Prof. dr. Jeroen Dewulf. In June 2018, she continued her PhD in the unit of Porcine Health Management of the same department under guidance of Prof. dr. Dominiek Maes, but in a new position as assisting academic staff. Her research focused on biosecurity and antimicrobial use in Belgian pig farms. She was also participating in the COST Action BETTER (Biosecurity Enhanced Through Training Evaluation and Raising Awareness) project (working group 1: mapping of biosecurity measures on farms and transport across Europe). Besides her research work, she conducted herd visits for the ambulatory swine clinic and for the Beter Leven label. She was also involved in teaching activities for second- and third-year Master students and supervised several students from the Faculty of Veterinary Medicine in completing their Master thesis. She gave several guest lectures and practical sessions for students of Animal Care and Agriculture (Bachelor in Agro- and biotechnology, HoGent) and Agricultural Sciences (Bachelor in Life Sciences, Ghent University).

Elise is first author and co-author of several scientific articles published in international peer-reviewed journals. Her work has been presented on different (inter)national conferences and meetings. In 2023, she obtained the certificate of the training program of the Doctoral Schools of Life Sciences and Medicine of Ghent University.

Currently, Elise is working at the Flanders Research Institute for Agriculture, Fisheries and Food (ILVO), where she supports the Pig, Poultry, and Cattle Information Centers.

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